

UNIVERSITY OF CAPE COAST

CHARACTERIZATION OF CASSAVA GERMPLASM
AND SCREENING FOR DIFFERENCES IN STARCH PRODUCTION
AND RESISTANCE TO CASSAVA MOSAIC VIRUS DISEASE

BY

PAUL AGU ASARE

CLASS NO.	
ACCESSION NO. 241323	
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SEPTEMBER 2010

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DECLARATION

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I hereby declare that this thesis is the result of my own original work and that no part of it has been presented for another degree in this university or elsewhere.

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We hereby declare that the preparation and presentation of the thesis were supervised in accordance with the guidelines on supervision of thesis laid down by the University of Cape Coast.

..... Date: 22/06/2011

PROF. JONATHAN PADI TETTEH

(PRINCIPAL SUPERVISOR)

..... Date: 22/06/2011

DR. ISAAC GALYUON

(CO-SUPERVISOR)

..... Date: 22/06/2011

DR. JUSTICE KWABENA SAFO

(CO-SUPERVISOR)

ABSTRACT

Cassava (*Manihot esculenta* Crantz) is a staple in sub-Saharan Africa. It is an important source of starch. Production is, however, constrained by many factors.

Forty-three accessions were assessed for morphological and genetic diversity using morphological descriptors and Simple Sequence Repeat (SSR) markers. The accessions were scored for their tolerance/resistance to the cassava mosaic disease. The properties of mixtures of cassava and sweet potato starch were also investigated.

Morphological descriptors revealed 4 clusters of accessions while Simple Sequence Repeat (SSR) markers revealed 9 distinct clusters of accessions, indicating a wider genetic diversity. Capevars was highly resistant to cassava mosaic virus disease, while most of the accessions were susceptible. Starch content was moderate, swelling volume, percentage solubility and swelling power were 14.5-35.3 ml g⁻¹, 8.3-27.5 % and 16.0-41.6 g g⁻¹ respectively. Maximum viscosity, breakdown and setback viscosity were 643.7 BU, 443.1 BU, and 141.9 BU. The pasting and peak temperatures of cassava starch increased with increase in the proportion of sweet potato starch. Set back viscosity was low for all accessions at a ratio of 20:80 cassava to sweet potato starch (C₂Sp₈).

SSR markers are recommended for the efficient management of germplasm and for effective utilisation of materials in breeding programmes to produce more resistant cultivars. Field screening for mosaic resistance should always be complemented with PCR or inoculation test. Genotype with good quality starch could be employed in the production of industrial starch.

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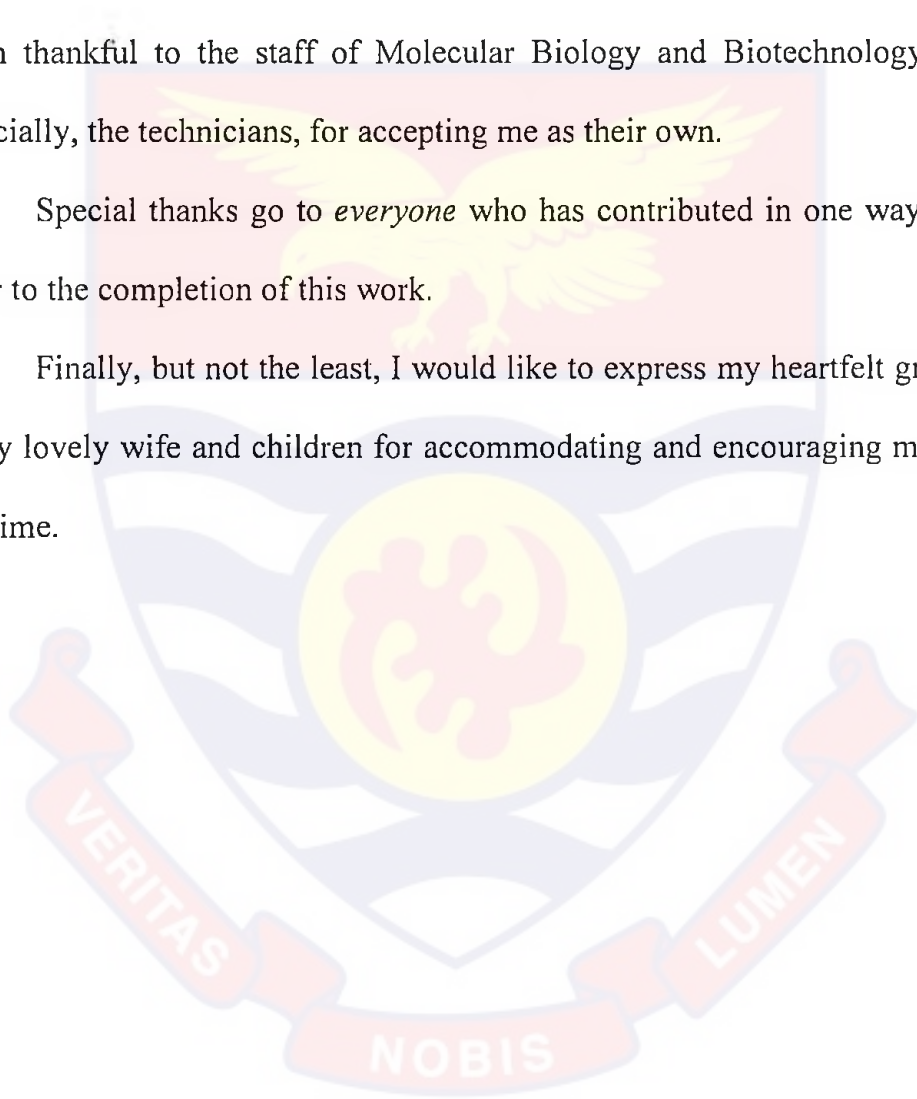
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DEDICATION

To my beautiful wife, Grace Evelyn, and our lovely kids, Mary Ann, Isaac

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LIST OF ABBREVIATIONS

ACMD	African cassava mosaic disease
ACMV	African cassava mosaic virus
AFLP	Amplified fragment length polymorphism
AGDP	Agricultural Gross domestic product
AGFW	Above ground fresh weight per plant
ANOVA	Analysis of variance
bp	Base pairs
BrL	Level of branching
BU	Brabender unit
BV	Brabender viscograph
°C	Degree Celsius
CAD	Cassava anthracnose disease
CBB	Cassava bacterial blight
CBSD	Cassava brown streak disease
cDNA	Complementary DNA
cpDNA	Chloroplast DNA
CGM	Cassava green mite
CIAT	International Centre for Tropical Agriculture
CM	Cassava mealybug
cM	Centimorgans
cm	Centimetre
cm ³	Cubic centimetre
CMD	Cassava mosaic disease
CMG	Cassava mosaic gemini

cmol	Centimolar
CRP	Colour of root pulp
CRI	Crop Research Institute
CRS	Colour of root surface
CTAB	Cetyltrimethylammonium bromide
CTS	Colour of tip shoots
CLL	Central leaf lobe length
CLW	Width of the central leaf lobe
CV	Coefficient of variation
DAP	Day after planting
DM	Dry matter
DNA	Deoxyribonucleic acid
DP	Degree of polymerization
dNTP	2'-deoxynucleoside 5'- triphosphate
EDTA	Ethylene-diaminetetraacetate
EACMV	East African Cassava Mosaic Virus
EACMV-Ug	East African Cassava Mosaic Virus-Ugandan variant
FAO	Food and Agricultural Organization
FBH	Height of first apical branch
FW	Fresh root weight
g	Gram
G	Genotype
GD	Genetic distance
HCN	Hydrocyanic acid
HFB	Height of first branch

HI	Harvest index
ICMV	Indian cassava mosaic virus
IITA	International Institute of Tropical Agriculture
IBPGR	International Board of Plant Genetic Resources
Kg	Kilogram
l	Litre
m	Metre
MAP	Months after planting
MAS	Marker-assisted selection
mg	Milligram
MgCl ₂	Magnesium chloride
MOFA	Ministry of Food and Agriculture
µg	Microgram
µl	Microlitre
min	Minute
ml	Millilitre
mm	Millimetre
ng	Nanogram
NTSYS	Numerical taxonomy multivariate analysis system
PC	Petiole colour
PCA	Principle component analysis
PCR	Polymerase chain reaction
PGRRI	Plant genetic resource research institute
PH	Plant height
PIC	Polymorphic information content

QTL	Quantitative trait loci
RAPD	Random amplified polymorphic DNA
RFLP	Restriction fragment length polymorphism
rpm	Revolution per minute
SACMV	South African Cassava Mosaic Virus
SC	Stem colour
SDW	Sterilized distilled water
SLCMV	Sri Lankan Cassava Mosaic Virus
SSR	Simple sequence repeat
Taq	<i>Thermus aquaticus</i>
TBE	Tris-borate EDTA
TE	Tris EDTA
Tris-HCl	Tris[hydroxymethyl]aminomethane hydrochloric acid
UPGMA	Unweighted pair group method with arithmetic averages
USDA	United States Department of Agriculture
UV	Ultraviolet
v/v	Volume per volume
WAP	Week after planting
w/v	Weight per volume
∞	Infinity

CHAPTER ONE

INTRODUCTION

Cassava or manioc (*Manihot esculenta* Crantz), a perennial shrub, currently is the sixth world food crop for more than 500 million people in tropical and sub-tropical Africa, Asia and Latin America (FAO, 2008). Cassava belongs to the Euphorbiaceae family containing 28 wild species and evolution from inter-specific hybridization among wild species. It is highly heterozygous due to its out-crossing nature. The DNA content is estimated around 1.7pg per cell nucleus with $2n = 36$ (Bennet et al., 1992).

Cassava (*Manihot esculenta* subsp. *esculenta*) is a staple crop with great economic importance worldwide (El-Sharkawy, 2003). It is one of the world's most important tropical plants, and is ranked as the fourth source of carbohydrates in the tropics (FAO, 1995). It is a staple crop in many developing nations in Africa, Asia and South America. Cassava is also an industrial crop for starch, flour and animal feed. It is particularly important to the poorest farmers because of its role in food security and as a source of income (Colvin et al., 2004). However, it is cultivated mainly by resource-limited small farmers for its starchy roots, which are used as human food either fresh when low in cyanogens or in many processed forms and products, mostly starch, flour, and for animal feed.

According to FAO (2008), the world's cassava production stands at 228.1 million tonnes with Nigeria as the largest cassava producing country

in the world, producing about 45.8 million tonnes, followed by Brazil with 27.3 million tonnes, Thailand 26.4 million tonnes, Indonesia 19.6 million tonnes, Ghana 9.7 million tonnes and India 7.6 million tonnes.

In Ghana, cassava is grown across all agro-ecological zones and ranks first in the area under cultivation and utilization; and also contributes 22 % of Agricultural Gross Domestic Product (AGDP). Over 70 % of Ghanaian farmers produce cassava and it is consumed by more than 80 % of the population of 22 million (Parkes, 2009).

The average yield of the crop in Ghana is 12.42 metric tonnes/ha against an achievable yield of 28.0 metric tonnes (MOFA, 2005). The inability of the crop to obtain its achievable yield is attributable to many factors. Chief among them is that farmers still use unimproved planting materials alongside few improved varieties which are seldom planted in pure stand. Secondly, farmers also use unimproved agronomic practices such as late planting, weed control at the wrong time, random planting and apathy in controlling diseases and pests. There is also little or lack of knowledge of the genetic diversity among the various accessions available for crop improvement.

Cassava, like any other crop, is affected by several factors which hinder or reduce its production. These factors are classified as biotic (disease, pest and weed attack) and abiotic (temperature, rainfall, solar radiation and photoperiod). Cassava mosaic virus disease is one of the major constraints to cassava production. The cassava mosaic virus disease (CMD), which is caused by any one or a combination of the white-fly-transmitted cassava mosaic begomoviruses, is an important constraint to

cassava production in Africa (Geddes, 1990; Zhou et al., 1997). It is spread mainly through the use of infected planting material. Estimated total crop yield losses due to CMD on the African continent amounts to about US \$440 million per annum (Thresh et al., 1997). The most effective means of controlling CMD is by the development of resistant varieties. Thresh et al. (1997) have reported that the effective means of controlling CMD is the use of resistant varieties.

A broad collection of 429 accessions were made from 11 districts each from the Western and Central Regions of the South-West ecozone of Ghana (Ampong-Mensah, 2000). Accessions were assembled and characterized based on morphology at the University of Cape Coast, and PGRRI at Bunso. All the 429 entries at UCC were found, after classification, to belong to 34 clusters (Amenorpe, 2002). The characterization was however based on morphological characters only.

For a meaningful crop improvement to be carried out, the true identity of the various cultivars available has to be established. Gepts (1993) has argued that molecular markers provide more useful information than morphological markers for studies on domestication and evolution of plants. Therefore, molecular characterization should be carried out to group cassava accessions in true breeding types for possible hybridization (Amenorpe, 2002). An understanding of the genetic diversity of cassava through the use of molecular markers would be important for guiding parental choice in breeding programmes and validating core collections (Hershey et al., 1994). Furthermore, knowledge of the genetic resources of

crop plants is important for facilitating the transfer of useful genes and maximizing the use of germplasm (Danquah et al., 2000).

With the increasing importance worldwide of cassava for food, animal feed and industrial use, there is an increasing need for a wide range of genetic resources to develop cultivars with specific characteristics and adaptation to different agro-ecologies. Furthermore, export demand for cassava in the form of chips has increased in the past years. In addition, the Government of Ghana has launched a special initiative to promote the production of starch for industrial purposes since 2001. All these events have excited interest in the expansion of cassava production. To achieve all these goals, there is the need to select and breed for improved cassava varieties.

Statement of the problem

There have been several collections of cassava germplasm. These collections have been grouped or characterized based on morphological features or descriptors. The assessment of phenotype may not be a reliable measure of genetic difference because of the influence of environment on gene expression. The analysis of plant DNA allows the direct assessment of variation in genotype. This is because DNA is not affected by the environment. Hence, molecular characterization of crops is more stable and reliable than characterization based on morphology (Gepts, 1993).

Recent evaluations of local accessions have revealed that some are promising in terms of tuber yield, starch yield and tolerance to cassava mosaic disease. Currently, the government special initiative on cassava is at

the verge of collapse. This is basically because the variety which is being used now (Afisiafi) has starch yield of just 18 %. This means that the factory has to process large quantities of tubers to get small amount of starch, which probably increases cost of production. Moreover, because of the low starch yield the company pays farmers a lesser amount of money in order to avoid serious losses. The farmers are unable to break even with the price they receive, hence the apathy to feed the company. Therefore, there is need to identify high-yielding cassava varieties for starch to enhance the growth of the cassava starch industry.

Justification

Characterization helps to get rid of duplicates and also saves resources and space in terms of storage. However characterization based on morphological traits is not reliable because they are influenced by the environment even though it is fast and good for preliminary evaluation. The characterization based on DNA or molecular markers is stable and reliable and will give a true distinction between the cassava lines available. Knowledge of relationships among lines is essential to the breeder, since it directs the exploitation of germplasm in hybrid production. The identification of promising local accessions would improve production and productivity of cassava. It will sustain the starch industry, generate income for small scale holders and reduce rural poverty. Starch of cassava with its high quality is suitable for specialty uses in food processing, textiles, and paper industries. It has excellent thickening and textural qualities, forms a clear gel, has good gel stability, a low tendency to retrograde, and has low

or no protein contamination (Ellis et al., 1998). The country stands to generate more export revenue from cassava chips and industrial starch as food security would be assured by expansion in cassava production to meet local and foreign markets, if new improved varieties are developed for farmers.

Statement of the objectives

Genetic diversity is the fuel for breeding. Currently, many varieties of cassava abound and several have various local names. The use of local names could pose a challenge since the same cassava variety could be named differently. Hence, there is the need to use molecular techniques to assess the cassava germplasm in Ghana to eliminate ambiguity. Furthermore, there is the need to identify cassava varieties with promise for breeding and selection for high yielding cultivars to sustain the cassava starch industry. Therefore, the main objective of the study was to assess the genetic diversity among the various accessions of cassava and identify promising ones based on starch content and viral resistance.

Specific objectives

The specific objectives of the study were to:

- characterize the accessions based on morphological traits as a preliminary evaluation;
- characterize selected number of accessions using SSR markers;
- identify mosaic-resistant genotypes;
- identify the various strains of cassava mosaic virus;

- identify high starch-yielding genotypes with excellent functional and pasting properties; and
- determine the effect of sweet potato starch on the functional and pasting properties of cassava starch as a physical modification method.



CHAPTER TWO

LITERATURE REVIEW

This chapter reviews the relevant research papers and other literature that are applicable to the topic. It will cover origin and distribution of cassava, botany of cassava plant, cassava production in Ghana, factors affecting cassava production, cassava utilization in Ghana, and variability of characters in cassava. Other aspect will be: the association among characters in cassava, the use of genetic markers in cassava improvement, the effect of mosaic virus disease on chlorophyll content of cassava, and cassava starch.

Botany of cassava

Taxonomy

Cassava (*Manihot esculenta*, Crantz) belongs to the class Dicotyledoneae, sub class Archichlamydeae, order Euphorbiales, family Euphorbiaceae, which comprises 7200 species. The Euphorbiaceae includes several commercially important plants, such as rubber trees (*Hevea brasiliensis*), oil plants (*Ricinus comumis*), root crops (*Manihot* spp.) and ornamental plants (*Euphorbia* spp.). Cassava belongs to the subfamily *Manigotae*, genus *Manihot*, and species name is *Manihot esculenta* Crantz.

Cassava is usually diploid, ($2n=36$). However, natural hybridization sometimes results in polyploid plants, such as triploids ($2n=3x=54$). Triploid and tetraploid plants differ from diploid plants in plant vigor, leaf shape and leaf size. Triploid plants usually grow and yield better than tetraploid and diploid plants (Ekanayake, et al., 1997).

The genus has two edible species, *M. utilissima* Phol (bittercassava) and *M. palmate* var. *api*, Phol (sweet cassava) based on high and low cyanogenic concentration, respectively (O'Hai, 1989). More recently it has been found that there is only one edible species and that is *M. esculenta*. It is the only one of 98 species of the family that is widely cultivated for food production.

Plant morphology

Roots

Roots are the main storage organs in cassava. In plants propagated from true seeds, a typical primary tap root system is developed, similar to dicot species. The radicle of the germinating seed grows vertically downward and develops into a taproot, from which adventitious roots originate. Later, the taproot and some adventitious roots become storage roots.

In plants grown from stem cuttings the roots are adventitious and they arise from the basal-cut surface of the stake and occasionally from the bud under the soil. These roots develop to make a fibrous root system. Only between 3 and 10 fibrous roots start to bulk and become storage roots. Most of the other fibrous roots are for nutrient absorption. Once a fibrous root

becomes a storage root, its ability to absorb water and nutrients decreases considerably. The storage roots result from secondary growth of the fibrous roots; thus the soil is penetrated by thin roots, and their enlargement begins only after the penetration has occurred.

Anatomically, the cassava root is not tuberous, but a true root, which cannot be used for vegetative propagation. The mature cassava storage root has three distinct tissues: bark (periderm), peel (or cortex) and parenchyma. The parenchyma, which is the edible portion of the fresh root, comprises approximately 85 % of total weight, consisting of xylem vessels radially distributed in a matrix of starch-containing cells (Wheatley & Chuzel, 1993).

The peel layer, which is comprised of sclerechyma and phloem, constitutes 11-20 % of root weight (Alves, 2002). The periderm, 3 % of the total weight, is a thin layer made of a few cells thick and, as growth progresses, the outermost portions usually slough off. Root size and shape depend on cultivar and environmental conditions; and size variability within a cultivar is greater than that found in other root crops (Wheatley & Chuzel, 1993).

Stems

The mature stem is woody, cylindrical and formed by alternating nodes and internodes. On the nodes of the oldest parts of the stem, there are protuberances, which are the scars left by the plant's leaves. A plant grown from stem cuttings can produce as many primary stems as there are

viable buds on the cutting. In some cultivars with strong apical dominance, only one stem develops.

The cassava plant has sympodial branching. The main stem (s) divides di-, tri- or tetra- chotomously, producing secondary branches that produce other successive branching. These branching, which are induced by flowering, have been called reproductive branching.

Stem morphological and agronomic characteristics are very important to characterizing a cultivar. The variation of these characteristics depends on cultivar, cultural practice and climatic conditions.

Leaves

Cassava leaves are simple, formed by the lamina and petiole. The leaf is lobed with palmated veins. There are generally an uneven number of lobes, ranging from one to nine (occasionally 11). Only a few cultivars are characterized by having three-lobed mature vegetative leaves, which may represent the primitive ancestral form (Rogers & Appan, 1973). Leaves near the inflorescence are smaller in size with fewer lobes (most frequently three-lobed), but the one closest to the base of the inflorescence is frequently simple and unlobed.

Leaves are alternate and have a phyllotaxy of 2/5, indicating that from any leaf (leaf 1) there are two revolutions around the stem to reach the sixth (leaf 6) in the same orthostichy as leaf 1. In these two revolutions there are five successive intermediate leaves (not counting leaf 1).

Mature leaves are glabrous and each leaf is surrounded by two stipules (approximately 0.5-1.0 cm long), which remain attached to the

stem when the leaf is completely developed (CIAT, 1984). The petiole length of a fully opened leaf normally varies from 5 to 30 cm, but may reach up to 40cm.

The upper leaf surface is covered with a shiny, waxy epidermis. Most stomata are located on the lower (abaxial) surface of the leaves: only a few can be found along the main vein on the upper (adaxial) surface (Alves, 2002). Of 1500 cultivars studied, only 2 % had stomata on the adaxial surface (El-Sharkawy & Cock, 1987). The stomata on the upper surface are also functional and bigger than those on the undersurface. Both are morphologically paracytic, with two small guard cells surrounded by two subsidiary cells. The number of stomata per leaf area range from 278 to 700 mm⁻², and all stomatal pores can occupy from 1.4 to 3.1 % of the total leaf area (Alves, 2002).

Flowers

Cassava is a monoecious species producing both male (staminate) and female (pistillate) flowers on the same plant. The inflorescence is generally formed at the insertion point of the reproductive branchings. Occasionally, inflorescences can be found in the leaf axils on the upper part of the plant. The female flowers, located on the lower part of the inflorescence, are fewer in number than male flowers, which are numerous on the upper part of the inflorescence. On the same inflorescence, the female flowers open 1-2 weeks before the male flowers (protogyny). Male and female flowers on different branches of the same plant can open at the

same time. Normally, cassava is cross-pollinated by insects; thus it is a highly heterozygous plant.

The flowers do not have a calyx or corolla, but an indefinite structure called perianth or perigonium, made up of five yellow, reddish or purple petals. The male flower is half the size of the female flower. The pedicel of the male flower is thin, straight and very short, while that of the female flower is thick, curved and long. Inside the male flower, there is a basal disk divided into ten lobes. Ten stamens originate from between them. They are arranged in two circles and support the anthers. The five external stamens are separated and longer than the inner ones, which join together on the top to form a set of anthers. The pollen is generally yellow or orange, varying from 122 to 148 μm in size, which is very large compared to other flowering plants (Alves, 2002). The female flower also has a ten-lobed basal disk, which is less lobulated than the male flower. The ovary is trilocular with six ridges and is mounted on the basal disk. The three locules contain one ovule each. A very small style is located on top of the ovary, and a stigma with three undulated, fleshy lobes originates from the style.

Fruit and seeds

The fruit is a trilobular capsule, ovoid or globular, 1-1.5 cm in diameter and with six straight, prominent longitudinal ridges or aristae. Each locule contains a single carunculate seed. The fruit has a bicidal dehiscence, which is a combination of septical and loculicidal dehiscences, with openings along the parallel plane of the dissepiments and

along the midveins of the carpels, respectively. With this combination of dehiscences, the fruits open into six valves causing an explosive dehiscence, ejecting the seeds some distance (Rogers, 1965). Fruit maturation generally occurs 75-90 days after pollination (Alves, 2002). The seed is ovoid-ellipsoidal, approximately 10mm long, 6 mm wide and 4 mm thick. The weight varies from 95 to 136 mg per seed (Alves, 2002). The smooth seed coat is dark brown, mottled with grey. The seeds usually germinate soon after collection, taking about 16 days for germination.

Origin and classification

Origin

Cassava (*Manihot esculenta* subsp. *utilissima*) is a staple crop with great economic importance worldwide, yet its evolutionary and geographical origins have remained unresolved and controversial. The genus *Manihot* (comprising 98 species) (Rogers & Appan, 1973) is distributed across much of the Neotropics, and the identity of cassava's closest wild relatives within the genus has been a source of widespread speculation (Renvoize, 1972; Rogers & Appan, 1973; Allem, 1994). Most traditional domestication hypotheses have envisioned the crop to be a "compilospecies" derived from one or more species complexes, either in Mexico and Central America (Rogers, 1965), and Rogers and Appan, (1973), or throughout the Neotropics (Sauer, 1993; Rogers, 1963; Ugent, et al., 1986). More recently, wild populations of *M. esculenta* that are likely to be the crop's direct progenitors have been identified in South America (Allem, 1987; Allem, 1994).

Different areas have been proposed to be the origin of cassava.

Records indicate that cassava might have been cultivated in Mayan Indian of Southern Mexico and Guatemala (Lundell, 1939). Reicche-Dolmatoff (1957), reported that the crop was cultivated in Venezuela and Colombia between 3000 and 7000 years ago.

Rogers (1963) postulated that cassava plant originated in North East Brazil with the likelihood of an additional centre of origin in Central America, Western and Southern Mexico and parts of Guatemala. He argued further that the crop might have been grown in Peru and Mexico 4000 and 2000 years ago, respectively. Reports by Rogers (1965) indicate that *M. awsculifolia* and *M. pringlei* which are wild relatives of cassava, are found wild in the areas of intensive cultivation which closely coincide with the areas of influence by the Mayan Indian civilization. Again, desiccated plant materials identified as cassava and dated as far back as 2500 BC have been found in Mexico (Roger, 1965).

Doku (1969) indicated that cassava is a native of South America, its centre of origin probably being Brazil, where one traveller enumerated nine cultivated races as early as 1637. Renvoize (1972), on his part, proposed that sweet and bitter cassavas have Mesoamerica and Venezuela as their first area of domestication respectively. Guatemala and Mexico, the Coastal Savannah of North-Western South America were also suggested as possible areas of origin (Rogers & Appan 1973). Purseglove (1987) also quoted Western and Southern Mexico and part of Guatemala and North-Eastern Brazil as the two geographical centres of speciation of the genus *Manihot*.

There is evidence of hybridization between cultivars and local natives resulting in the formation of complexes from which the wild species, *M. saxicola* Lang. was believed to have been derived. Nassar (1978) and Lancaster et al. (1982) gave northern Amazonian region and northern South America as areas of domestication of the crop.

Comparative studies of reproductive characteristics, botanic origin and phylogenetic relatedness of cassava with other *Manihot* species, as well as the history of the domestication of cassava are still in the early stages. Oslen and Schaal (1999) investigated cassava domestication in a phylogeographic study based on the single-copy nuclear gene glyceraldehyde 3-phosphate dehydrogenase (*G3pdh*). The *G3pdh* locus provides high levels of noncoding sequence variation in cassava and its wild relatives, with 28 haplotypes identified among 212 individuals (424 alleles) examined. These data represent one of the first uses of a single-copy nuclear gene in a plant phylogeographic study and yielded several important insights into cassava's evolutionary origin which include:

- (i) cassava was likely domesticated from wild *M. esculenta* populations along the southern border of the Amazon basin;
- (ii) the crop does not seem to be derived from several progenitor species, as previously proposed; and
- (iii) cassava does not share haplotypes with *Manihot pruinosa*, a closely related, potentially hybridizing species.

These findings provide the clearest picture to date on cassava's origin. When considered in a genealogical context, relationships among the *G3pdh* haplotypes are incongruent with taxonomic boundaries, both within *M.*

esculenta and at the interspecific level. This incongruence is probably a result of lineage sorting among these recently diverged taxa. Although phylogeographic studies in animals have provided many new evolutionary insights, application of phylogeography in plants has been hampered by difficulty in obtaining phylogenetically informative intraspecific variation. Hence, their study demonstrates that single-copy nuclear genes can provide a useful source of informative variation in plants.

In sum, cassava can be said to be a new world crop with its origin in South America which includes Brazil, Venezuela, Peru, Bolivia and parts of Argentina and Mesoamerica which includes Mexico, Guatemala, Maya India, El Salvador and Nicaragua.

Classification

Morphological classification

Cassava can be classified based on distinctive morphological features such as leaf size, colour and shape, branching habit, plant height, colour of stem and petiole, tuber shape and colour, time to maturity and yield. Of these traits, branching habit and time to maturity are more commonly used.

Cassava varieties can be grouped into branching and non-branching. These branching types can be divided into early and medium branching. Ekanayake et al. (1997) recorded some early branching types as low as 20 cm. Profuse branching types have a higher yield potential, leaf number, and leaf area index than other types. Non-branching types required closer plant spacing than branching types to achieve similar levels of leaf area index

and yield (Msikita et al. 2000). Plants with only one or two stems tend to branch more profusely, thereby compensating for the fewness of stems or when interplant spacing is very wide (Onwueme, 1982).

Cassava can be classified into short-season and long-season accessions. Short-season accessions mature as early as six-months after planting while long-season accessions take at least a year to mature and once matured have to be harvested else they deteriorate (Purseglove, 1987).

Biochemical classification

Early classification of cassava placed them into two distinct types bases on hydrocyanic level – bitter and sweet cassava. The bitter varieties belong mainly to *M. esculenta* (syn *M. utilisima*) while the sweet varieties are mainly *M. palmate* (syn *M. dulcis*).

The sweet varieties have hydrocyanic acid (HCN), with the HCN confined to the phelloderm of the tuber, whereas the bitter ones have high HCN content generally distributed everywhere in the tuber (Purseglove, 1987). The sweet bitter distinctions are not justified as both *M. esculenta* and *M. palmate* have bitter and sweet varieties and the hydrocyanic level of an accession also depends on the environment (Ciferri, 1938; Purseglove, 1987). Dimpleby and Ucko (1969) have also argued that there is no relationship between morphological features of the plant and its HCN level but rather the HCN levels vary with soil and climatic conditions as well as age of the plant.

The IITA (1990) classification also based on quantity of HCN per 100g fresh tuber weight, groups cassava varieties as having high HCN

varieties, ($\geq 10\text{mg}$), intermediate HCN varieties, (5-10mg) and low HCN varieties ($< 5\text{mg}$).

Some caution should be exercised in using the levels of glucoside as distinguishing characteristics for cassava cultivars. This is because the exact level of glucoside in a particular cultivar will vary according to the environmental conditions under which the plant is grown (Onwueme, 1978).

Doku (1969) has also argued that taste is subjective and there is no clear cut demarcation between sweet and bitter varieties because of the existence of several intermediate types. HCN levels are also known to increase with drought, high soil fertility and potassium deficiency. Furthermore, bitter and sweet tubers could be harvested from the same plant and in some cases sweet varieties tend to have more HCN than the bitter cultivars (Doku, 1969). Hence, such classification cannot be relied upon for the identification of cassava varieties.

Molecular classification

The most recent method of classification of crop plants that has gained root in crop improvement is the classification based on the genetic makeup of the plant. This is because it saves time and the results are reliable. Gepts (1993) argues that molecular markers provide more useful information than morphological markers for studies on domestication and evolution of plants. Thus, in addition to morphological characterization, molecular characterization, based mainly on DNA molecular markers, has been very useful in order to evaluate germplasm genetic diversity

(Beeching et al., 1993; Fregene et al., 1994). An understanding of the genetic diversity of cassava through the use of molecular markers would be important for guiding parental choice in breeding programs and validating core collections (Hershey et al., 1994). Therefore, it has been argued that molecular characterization should be used to classify cassava accessions into true breeding types for possible hybridization (Amenorpe, 2002).

Several genetic markers have been developed for classification of cassava germplasm into various clusters. Classification based on the genetic makeup of the plant is important and has been applied in cassava using diverse number of molecular markers. For instance, RAPDs (Tonukari et al., 1997; Carvalho & Schaal, 2001) and SSR (Carvalho & Schaal, 2001) have been used for the identification of close relatives of cassava. Furthermore, RAPDs (Angel et al., 1992; Colombo et al., 2000; Asante & Offei, 2003), AFLP (Colombo et al., 2000; Elias et al., 2001), RFLP (Angel et al., 1992) and SSR (Fregene et al., 2001) have been used for the characterization of germplasm and development of large numbers of new varieties with diverse agronomic traits.

Growth and development

Plant developmental stages

Cassava is a perennial shrub, so it can grow indefinitely alternating periods of vegetative growth, storage of carbohydrates in the roots, and even periods of almost dormancy, brought on by severe climatic conditions such as low temperature and prolonged water deficit. Bigger plants tend to yield bigger root tubers, since there is a positive correlation between the

total biomass and storage root biomass (Ramanujam, 1990). During its growth, there are distinct developmental phases. The occurrence, duration and existence of each phase depend on the variety, environmental conditions and cultural practices. The periods and main physiological events during the growth of a cassava plant under favourable conditions in the field are summarized below:

Emergence of sprouting usually occurs 5-15 days after planting (DAP). From 5-7 DAP the first adventitious roots arise from the basal cut surface of the stake and occasionally from the buds under the soil. During the 10-12 DAP the first sprouting occurs, followed by small leaves which start to emerge (Alves, 2002). Emergence is achieved at 15 DAP.

The beginning of leaf development and formation of root system occur 15-90 DAP. The true leaves start to expand around 30 DAP when the photosynthetic process starts to contribute positively to plant growth. Until 30 DAP, shoot and root growth depends on the reserves of the stem cutting. The fibrous roots start to grow, replacing the first adventitious roots. These new roots start to penetrate in the soil, reaching 40-50 cm deep, and function in water and nutrient absorption (Alves, 2002). Few fibrous roots (between three and 14) will become storage roots, which can be distinguished from fibrous roots from 60 to 90 DAP (Alves, 2002). At 75 DAP the storage roots represent 10-15% of total dry matter.

Canopy establishment starts from 90-180 DAP. Maximum growth rates of leaves and stems are achieved in this period, and the branching habit and plant architecture is defined. From 120 to 150 DAP the leaves are able to intercept the most of the incident light on canopy (Velkamp,

1985). Maximum canopy size and maximum dry matter (DM) partition to leaves and stems are accomplished (Howeler & Cadavid, 1983; Ramanujam, 1990; Alves, 2002). The storage root continues to bulk. The most active vegetative growth for cassava occurs in this period (Alves, 2002).

High carbohydrate translocation to roots occurs 180-300 DAP. Photoassimilate partition from leaves to roots is accelerated, making the bulking of storage roots faster. The highest rates of DM accumulation in storage roots occur within this period. Leaf senescence increases, hastening rate of leaf fall and stem becoming lignified (Alves, 2002).

Finally, dormancy starts from 300-360 DAP. Rate of leaf production is decreased. Almost all leaves fall and shoot vegetative growth is finished. Only translocation of starch to root is kept, and maximum DM partition to the roots is attained. This phase occurs mainly in regions with significant variation in temperature and rainfall. The plant completes its 12-month cycle, which can be followed by a new period of vegetative growth, DM accumulation in the roots and dormancy again (Alves, 2002).

Cassava Introduction and Production in Ghana

In Africa, cassava was first introduced to the Congo from South America about 400 years ago (Doku, 1969). Currently, cassava is cultivated in about 40 African countries, stretching through a wide belt from Madagascar in the Southeast to Senegal and to Cape Verde in the Northwest (Nweke, 2004). Throughout the forest and transition zones of Africa, cassava is either a primary staple or a secondary food staple.

Cassava has been grown in Ghana since 1750 (Doku, 1969). It is the most favoured among all the root crops and indeed all food crops eaten by the Ghanaian consumer (Annor-Frimpong, 1994).

Cassava transformation in Ghana has lagged behind Nigeria by about a decade. For example, the dramatic increase in cassava production occurred in Nigeria from 1984 to 1992 and in Ghana from 1990 to 2001. Until the drought which occurred in the early 1980s and resulted in the failure of food crops except cassava in Ghana, government agricultural policies emphasized on large scale production of grains by the public sector and neglected cassava as an inferior food whose consumption was destined to decline as incomes increased. Ghana, the seventh largest producer in Africa in the early 1960s, with an annual production of only 1.2 million tons, increased its output six-fold and, by the late 1990s, produced 7.2 million tons annually and advanced to the position of the third largest producer in Africa after Nigeria and the Congo (Nweke, 2004)

The crop was first introduced in the Volta Region and from there it spread to parts of the Ashanti and Brong-Ahafo Region in the forest belt. It is now grown in all the regions, except the Upper East (MOFA, 2000). The slow rate of spread following its introduction was probably due to the slow dissemination of knowledge about the various methods of preparing foods from it (Doku, 1969). Although cassava seems to have been absent in Ghana at the beginning of the 18th century, it was widely cultivated around Accra in 1785 (Carter et al., 1995). Cassava became a major food crop in Ghana on the coastal plains at the beginning of the 19th century, and reached Ashanti and the North during the early 1930s (Doku, 1969).

The bulk of the nation's cassava is produced in the south and middle of Ghana, which accounts for roughly 80% of the total cassava production in Ghana. In the Northern Region of Ghana only 20% is devoted to cassava production; however, the production and area under cultivation is increasing as cassava also grows on marginal land and yields a sufficient amount for the semi-subsistence farmers, who often lack cash for fertilizer or other expensive inputs for crop production.

Currently, cassava plays an important role in food security in Ghana. The Ewes have named the crop *agbeli*, which means *there is life*. This name no doubt portrays its importance to the whole country and to the Ewes, in particular, who are not only the major cassava growers but also producers of gari, starch, tapioca, kokonte and many other products made from cassava (Doku, 1969).

In 2002, Ghana's cassava yield stood at 12,249kg ha⁻¹, which was higher than that of Nigeria, 9979 kg ha⁻¹ (FAO, 2002). Total production for same year was 34476 million tonnes and 9731 million tonnes for Nigeria and Ghana, respectively (FAO, 2002).

Factors affecting cassava production

Biotic, abiotic and agronomic factors affect the production and yield of cassava.

Biotic factors

The biotic factors that affect cassava production in Africa include diseases, pests, and weeds. These constraints have contributed to keeping

the average cassava yield in Africa at 6.4 t ha^{-1} , which is well below the world average of 8.8 t ha^{-1} (IITA, 1990). Diseases alone can cause as high as 90% reduction in tuber yield (Lozano & Booth, 1994).

Diseases

Fungal diseases

Most fungal diseases of cassava are soil borne and cause root rot. There are few of them which cause damage to the leaves. At the early stages of growth they cause damping off. *Sclerotium* rot, caused by *Sclerotium rolfsii*, is the most common tuber rot disease and occurs on roots and tubers at all stages of development (IITA, 1990). It can be recognized by the appearance of white mycellial growth on infected roots and tubers (IITA, 1990). This disease can be prevented by rotating cassava with other crops and avoiding recently drained plains where the water table is too close to the soil surface (Janssens, 2001).

Anthraxnose, caused by *Colletotrichum gloeosporioides*, causes brownish spots on the leaves. Necrotic lesions may appear on stems, branches and petioles (Janssens, 1998). Cassava anthracnose disease (CAD) is the most important stem disease in Africa and it occurs in all major cassava-growing areas.

Armillaria mellea and *Fomes lignosus* are other soil-borne fungi which may cause root rot and kill cassava plants in patches. The problem is prevalent particularly on newly cleared forest land. The damage can be contained by complete uprooting of the affected plants.

Another fungal disease of cassava is cercospora leaf spot. It affects the leaf of cassava. There are three types of cercospora leaf spot; the most common being brown leaf spot caused by *Cercosporidium henningsii* (IITA, 1990). The other types are leaf blight, caused by *Cercospora vicosacs*, and white leaf spot caused by *Cercospora caribac*. They are not known to kill plants, because symptoms are restricted to older leaves and set in after tuberization has occurred (IITA, 1990).

Bacterial diseases

Cassava bacterial blight (CBB) is the most wide-spread bacterial disease of cassava and is second in importance only to the Africa cassava mosaic virus in Africa (IITA, 1990). It is caused by *Xanthomonas compestris pathovar manihot*. The symptoms include characteristic angular water soaked leaf spot, blight, gum exudation, stem die back, wilt and vascular necrosis (IITA, 1990; Janssens, 2001).

Viral diseases

The best-known viral disease of cassava is mosaic caused by gemivirus. It is transmitted from one cassava plant to another plant by the white fly, *Bemisia tabaci*. It is also spread between plantations and from one region to another by the use of infected planting materials. Symptoms include characteristic light green, yellow to white patches, irregularly intermingled. The chlorotic areas may be only small flecks or spots or they may cover the entire cassava leaf. The mottling is sometimes accompanied by leaf deformation and a general stunting of the plant.

Another viral disease of cassava is the cassava brown streak virus disease. The virus destroys the root while the leaves appear to be healthy, so farmers do not realize that their crop has been ruined until harvest time (Kanju, 2007).

Cassava mosaic disease

Cassava mosaic disease (CMD) caused by mosaic gemini viruses (*Geminiviridae: Begomovirus*) is undoubtedly the most important constraint to the production of cassava in Africa at the outset of the 21st century (Braima et al., 2000). Although the disease was recorded for the first time in the latter part of the 19th century, for much of the intervening period it has been relatively benign in most of the areas where it occurs and has generally been considered to be of minor economic significance (Braima et al., 2000). African cassava mosaic disease was first observed near Accra in 1926 (Doku, 1966) and its spread was more significant in the coastal areas of the country around 1930 (Leather, 1959; Clerk, 1974). At present, the disease is widespread and is found in all the agro-ecological zones in Ghana. Although in Ghana yield losses due to ACMV have not been quantified, the importance of this disease is indicated by leaf chlorosis, distortion of leaves and stunting of plants. Losses due to ACMV disease range from 20 to 95 per cent (Beck & Chant, 1958; Fargette et al., 1988; Hahn et al., 1989; Terry and Hahn, 1990; Otim-Nape et al., 1994; Braima et al., 2000).

CMD continues to be prevalent in all the main cassava-growing areas in Africa, and is generally regarded as the most important disease,

causing between 20 and 90 % crop losses depending on the cultivar, viral strain and environmental factors. Environmental factors favouring the development and fecundity of the white fly (vector) enhance the spread of the disease. The mosaic virus spread is therefore highly linked with its whitefly vector (Fargette, et al., 1985). The lower leaves of the infected plants look apparently healthy while those above the point of first infection show severe symptom expression, drastic reduction in leaf size with marked distortion. The plants harbour numerous adult white fly populations on the young shoots and large nymph populations on the lower surface of the apparently healthy lower leaves. Lack of alternative propagation stock in the disease-infected areas provides farmers no choice but to use material from the previous harvest of infected plants, thereby spreading the disease season by season.

Studies have, however, shown that the effects of ACMV on yield depend on variety and stage of infection, but are usually substantial as indicated above. Plants grown from infected cuttings are much more seriously affected than those infested later by the whitefly vector (*Bemisia tabaci*) and plants infected at a late stage of crop growth are almost unaffected. Positive relationships have been established between the extent and severity of the leaf symptoms and yield loss, but losses can be considerable, even in varieties designated as resistant (Thresh et al., 1994).

Based on recent taxonomic guidelines, several strains of cassava mosaic virus have been identified (Fauquet & Stanley, 2003), but given the apparent propensity of the cassava mosaic gemiviruses (CMGs) to exchange genetic material, this should be viewed as a dynamic tool to aid

those working with these organisms rather than a fixed system. Based on this new approach, in which the sequence homology demarcation between species has been set at 89% for the DNA-A component of begomoviruses (Fauquet & Stanley, 2003), six African and two Indian CMG species are recognized. These are the Africa Cassava Mosaic Virus (ACMV), EACMV, East African cassava mosaic Cameroon virus (EACMCV), East African cassava mosaic Malawi virus (EACMMV), East African cassava mosaic Zanzibar virus (EACMZV), East African cassava mosaic virus-Ugandan variant (EACMV-Ug), South African Cassava Mosaic Virus (SACMV), Indian Cassava Mosaic Virus (ICMV) and Sri-Lankan cassava mosaic virus (SLCMV) (Fauquet & Stanley, 2003). Different viruses have very different biological characteristics often with gross differences in the severity of disease caused (Harrison et al., 1997; Fondong et al., 2000; Pita et al., 2001). There is an obvious potential economic advantage to be gained from understanding which viral strain, and strain combinations occur and how they are distributed.

Pests

Insects

There are at least six major insect pests of cassava in Africa. The cassava mealy bug, *Phenacoccus manihoti*, the variegated grasshopper, *Zonocerus variegatus*, the elegant grasshopper *Z. elegans*, the cassava scale insect, *Aonidomytilus albus*, the coreid bug, *Pseudotheraptus devastans* and the whitefly, *Bemisia tabaci*. Other pests include the stripped mealybug, *Phenacoccus madeirensis* (IITA, 1990).

Whitefly is the vector of ACMV, and is prevalent throughout Africa (Storey & Nicholas, 1938). The reproduction and activity of the whitefly are encouraged by high rainfall, a temperature range of 25 to 27°C and high light intensity (Fauquet & Stanley, 2003). Under field conditions, the spread of ACMV by whitefly occurs mainly in April, May and June when the population is high (IITA, 1990).

Mites

The most important cassava pests in Africa are cassava green spider mite (CGM) and red spider mite (RSM). Indigenous to South America, CGM was reported in forest Uganda in 1972 (Onwueme, 1978). It has since spread rapidly over much of Africa. Only one species is found on the continent, *Mononychellus tanajoa* (Onwueme, 1978). Cassava Green Mite sucks cell sap from leaf tissues. The damage first appears on the surface of developing and newly formed leaves. Symptoms vary from a few chlorotic spots to complete chlorosis and may be mistaken for ACMV symptoms (IITA, 1990; Janssens, 2001).

Nematodes

The nematodes *Meloidogyne* spp and *Rotylenchulus reniformis* cause damage to cassava roots. To date, no resistant variety has been identified. Planting the legume *Crotalaria* spp. during fallows period makes it possible to trap nematodes and thus reduce their attacks. Spreading cacao pod debris is also said to reduce the incidence of nematodes (Janssens, 2001).

Weed

Cassava can be seriously affected by early weed infestation. Slow initial growth and development make the plant susceptible to weed interference during the first 3 to 4 months after planting (IITA, 1990). Weed competition in cassava crops reduces canopy development, tuberization and tuber number. Reduction in tuber yields varies from 40 % in the early-branching cultivars to nearly 70 % in the late- or non-branching cultivars. The problem with some weeds, such as *Imperata cylindrica*, is not limited to direct yield reduction; this weed also causes mechanical damage to cassava tubers which provides a route of entry for fungi damage and other pathogens that cause tuber rot and reduce quantity of produce.

Abiotic factors

Cassava is found over a wide range of edaphic and climatic conditions between 30 °N and 30 °S latitude, growing in regions from sea level to 2300 m altitude, mostly in areas considered marginal for other crops. The most important abiotic factors that affect cassava production are temperature, photoperiod, solar radiation and rainfall.

Temperature

Temperature affects sprouting, leaf size, leaf formation, storage root formation and, consequently, general plant growth. The behaviour of cassava under the temperature variations that usually occur where cassava is normally cultivated indicates that its growth is favourable under annual

mean temperatures ranging from 25 to 29 °C, but it can tolerate from 16 to 38 °C (Alves, 2002).

At low temperatures (16 °C) sprouting of the stem cutting is delayed, and rate of leaf production, total and storage root dry weight is decreased. Sprouting is hastened when the temperature increases up to 30 °C but is inhibited with temperatures > 37 °C (Alves, 2002). As temperature decreases, leaf area development becomes slower because the maximum size of individual leaves is smaller, and fewer leaves are produced at each apex although leaf life is increased (Irikura et al., 1979). At a temperature of 15-24, the leaves remain on the plant for up to 200 days (Irikura et al., 1979), while at higher temperatures are 120 days (Alves, 2002).

There is a genotype-by-temperature interaction for yield ability. Irikura et al., (1979) evaluated four cultivars under different temperatures and found that higher yields were obtained at different temperatures according to the cultivar, indicating that the effect of natural selection is highly significant on varietal adaptation.

The main effect of temperature on biological production, such as DM partitioning does not change much when cassava is cultivated under different temperatures (Alves, 2002). Higher temperatures are associated with a greater crop growth rate (CGR) and high photosynthetic rate. El-Sharkawy et al., (1992) evaluated the potential photosynthesis of three cultivars from contrasting habits under different growing environments and observed that photosynthetic rate increased with increasing temperature, reaching its maximum at 30-40 °C. In all cultivars photosynthesis was substantially lower in leaves that had developed in the cool climate than in

those from the warm climate. The high sensitivity of photosynthesis to temperature points to the need for genotypes more tolerant to low temperature, which could be used in the highland tropics and subtropics.

Photoperiod

Day length affects several physiological processes in plants. The differences in day length in the tropical region are very small, varying from 10-12 h throughout the year. Thus photoperiod may not limit cassava root production in this region. On the other hand, the restrictions regarding cassava distribution outside the tropical zone can be due to effects of day length variation on its physiology. Although studies about day length effect in cassava are scarce, tuberization, photoassimilates partitioning and flowering are reportedly affected. Photoperiods may affect the hormonal balance in the plant; for instance, it may influence the level of Gibberellic Acid (GA₃) and Indole Acetic Acid (IAA). Besides, photoperiod interacts with temperature, especially night temperature, but varietal differences in the nature of the interaction are also found (IITA, 1990).

Experiments in which the day length was artificially changed have shown that the optima light period for cassava is around 12 h, with probable varietal differences in the critical day length (Alves, 2002). According to IITA, (1990) most varieties initiate tubers only under short-day conditions. Long days promote growth of shoots and decrease storage root development, while short days increase storage root growth and reduce the shoots, without influencing total dry weight (DW). The increase in shoot DW under long days is a result of significant increases in plant

height, leaf area per plant, number of apices per plant, and number of living leaves per apex (Veltkamp, 1985). This suggests an antagonistic relationship between shoot growth and development of the storage roots in response to variation in day length.

Solar radiation

The commonest cassava production system is intercropping with other staple crops. In Latin America and Africa, cassava is usually associated with an earlier maturing grain crop such as maize, rice or grain legumes (beans, cowpeas or groundnuts) (Mutsaers et al., 1993). Cassava is also intercropped with perennial vegetation (Ramanujam et al., 1984). Cassava is usually planted after the intercropped species. Even when it is planted at the same time, the associated crop such as maize is established faster than cassava. Thus in an associated cropping system cassava is always subjected to different degrees of shading and low light intensity in the early stages of development. Considering that cassava is a crop that requires high solar radiation to perform photosynthesis more efficiently (El-Sharkawy et al., 1992), it is very important to know the effect of shade on cassava development and production. Ramanujam et al. (1984) evaluated 12 cassava cultivars under the shade in a coconut garden (85-90% shading). Under shading, the root bulking process started about 3 weeks after that in plants grown without shading, and the number of storage roots per plant and NAR was reduced under shading. Okoli and Wilson (1986) submitted cassava to six shade regimes and observed that all levels of shade delayed

storage root bulking and at 20, 40, 50, 60, and 70 % shade reduced cassava yield by 43, 56, 59, 69 and 80 %, respectively.

In relation to shoots, under field conditions, shading increases plant height and the leaves tend to become adapted to low light conditions by increasing leaf area per unit weight (Ramanujam et al., 1984; Oloki & Wilson, 1986) and shortening leaf life only under severe shading. Under ideal growing conditions, cassava leaves have a life of up to 125 days (Alves, 2002). Levels of shade up to about 75 % have very little effect on leaf life, but under 95-100 % shade, leaves abscise within 10 days (Alves, 2002). Thus under limited photosynthesis caused by low solar radiation, most of the photosynthates are utilized for shoot growth, affecting storage root development significantly, showing that the shoots are a stronger sink than roots.

Agronomic practices

Cassava like any other crop requires good cultural practices to ensure high yield. Proper land preparation, right planting with good planting materials, weed control at the right time, intercropping and harvesting at the right time are some of the cultural practices that enhance high yield.

Traditional farmers seldom follow recommended cultural practices for cassava, and may be unaware of the existence of improved varieties. The use of unimproved varieties, together with inadequate length and age of planting material and incorrect plant population, depth and time of planting, are among the reasons why yields under most traditional systems

are low. The selection of good planting material is one of the most important aspects of cassava production; the material must be fresh and taken from healthy and mature stem portions if high yields are to be realized (IITA, 1990).

In traditional systems, land preparation starts before the onset of the rainy season and consists of clearing the vegetation and burning it. Mounds or ridges are made at the beginning of the rains. On sandy soils there is little land preparation; farmers merely slash weeds and plant cassava cuttings in relatively undisturbed soil. Cassava is mainly intercropped with yams, cocoyams, maize, melons and beans in a single plot i.e. polyculture. According to Andrews and Kassam, (1976) cassava can be used in alley cropping system with *Gyricidia sepium* and *Flemingia marcrophyll*. However it is known to reduce yield of cassava (Ekanayake, 1995).

Today, cassava is largely grown as a single crop, i.e. monoculture or with one other crop, i.e. diculture, because of the increasing need to grow cassava. This practice has increased the rate of soil erosion in cassava growing areas (Odemerho & Avwunudiogba, 1993).

One of the reasons for the widespread cultivation of cassava is the crop's ability to grow in soils that are too impoverished to support other staple crops. This is because the crop has an extensive root system and is able to utilize plant nutrients less accessible to other crops. Despite this advantage, the crop is a heavy feeder of nitrogen and potassium. Phosphorus, calcium and magnesium are, however, required in minimum amount. For instance a yield of 25 tonnes per hectare of the crop requires

122 kg N, 27 kg P, 145 kg K, 45 kg Ca and 20 kg Mg of nutrient (Addo-Quaye et al., 1993).

Cassava utilization in Ghana

Food uses

Cassava is consumed in a wide variety of forms. The importance of cassava in the world food supply is due to its durability as a plant and also due to it being a cheap and excellent source of dietary carbohydrate. In Ghana, cassava roots are usually prepared and eaten in the form of 'fufu', 'ampesi', 'agblima', 'akple', 'banku', and 'yakayeke'. The roots can be roasted and eaten and they can also be processed into dry chips (*kokonte*), *gari*, biscuits, buns and doughnuts, breads and cakes (MOFA, 2000). Only cassava tubers that give soft and friable texture (good cooking qualities) after boiling or roasting are suitable for fufu and ampesi.

Cassava can also be processed into dry chips (*kokonte*), tapioca, *gari*, fufu-flour, cookies, biscuits, buns, doughnuts, breads and cakes (MOFA, 2000). In many areas, the roots are consumed as a major staple, although in some places boiled fresh cassava roots are eaten as vegetable. In large parts of Africa, particularly Central Africa, the leaves are also consumed as a leafy vegetable (Dorosh, 1988).

Feed uses

Cassava is used as food for fattening farm animals such as pigs, cattle and poultry. It is fed to pig or poultry either raw or processed into chip (Quaye, 1979). On account of its low protein content, it is usually

blended with maize or mixed with protein concentrates such as oil seed cake and mineral salts. In the case of pigs, the performance was progressively better as the level of cassava feed was increased to 40%. In view of the potential value of cassava to supply energy to dairy cattle, it has been used in a great number of experiments as the main source of energy, resulting in higher milk and fat yields and live weight gains (Pineda & Rubio, 1972). Similar results have been obtained for beef cattle when steers fed on commercial concentrate and cassava-based diets. Better performance of bulls has also been reported by Montilla et al. (1975) on 40 % cassava rations rather than on maize meal.

The high nitrogen content of cassava leaves has attracted attention in animal feeding and, in some countries the making of cassava silage from whole plants is practiced. Cassava peels are also fed or dried to sheep and goat (Doku, 1969). Cassava roots are used as feed for farm animals usually to substitute for a part of the main ingredients in nutritionally balanced rations. For example, Gomez et al. (1984) in Colombia reported that when cassava was substituted for maize in a poultry broiler ration at levels of up to 30 %, there was no significant difference in the performance at all levels, but the 20 % level substitution was the most economical. It was noted that high levels of cassava intake were more acceptable for broiler production than for laying hens. Egg production and quality could be adversely affected by nutritional imbalances associated with rations high in cassava.

Industrial uses

Cassava has several industrial uses. It is a source of starch in the textile and paper industries. It is also used in the brewing and bakery industries (MOFA, 2000). The peel and stems find uses as fuel in the fishing industry along the coast of Ghana (Doku, 1969). Some of the industrial uses of cassava have been tabulated below.

Table 1: Non-food applications of cassava starches

Adhesive	Metals Industry
➤ Hot-melt glues	➤ Foundry core binder
➤ Stamps, book binding, envelopes	➤ Sintered metal additive
➤ Labels (regular and water proof)	➤ Sand casting binder
➤ Wood adhesives, laminations	Textile Industry
➤ Automotive, engineering	➤ Warp sizing
➤ Pressure sensitive adhesives	➤ Fabric finishing
➤ Corrugation	➤ Printing
➤ Paper sacks	Cosmetic and
Explosives Industry	Pharmaceutical Industry
➤ Wide range binding agent	➤ Dusting powder
➤ Match-head binder	➤ Make-up
Paper Industry	➤ Soap filler/extender
➤ Internal sizing	➤ Face creams
➤ Filler retention	➤ Pill coating, dusting agent
➤ Surface sizing	➤ Tablet binder/dispersing agent
➤ Paper coating (regular and colour)	Mining Industry
➤ Carbonless paper stilt material	➤ Ore flotation
➤ Disposable diapers, feminine products	➤ Ore sedimentation
Construction Industry	➤ Oil well drilling muds
➤ Concrete block binder	Miscellaneous
➤ Asbestos, clay/limestone binder	➤ Biodegradable plastic
➤ Fire-resistant wallboard	➤ Dry cell batteries
➤ Plywood/chipboard adhesive	➤ Printed circuit boards
➤ Gypsum board binder	Leather finishing
Paint filler	

Adopted from Adomako 2007.

Variability of characters in Cassava

Several variabilities have been reported in Africa among the local cassava germplasm collection. This could be attributed to the fact that some of the materials flower and set seed freely and new cultivars are established from volunteer seedlings. Because cassava is a cross-pollinated crop, continuing recombination or variation occur from outcross of genetically heterozygous cultivars (IITA, 1990). Secondly, spontaneous mutation may give rise to additional genetic variation although this has been proven to be rare. Consequently, large variability has been reported within *M. esculenta* for several agronomically important characters including leaf, stem and root characters, resistance to pest and diseases, root yield (CIAT, 1973, 1975, 1976), abiotic stress tolerance, adaptability to the environment and product quality (Ogunbodede, 1997).

The essential traits that constitute the storage root yield are root weight, root number, and root size. Storage root weight is dependent on both, genetic and environmental factors such as temperature, soil conditions, rainfall and photoperiodism that influence the physiological process that affect the yield of cassava plant (Cock, 1983). Genetically, storage root yield has been reported to be associated with traits like profuse branching, and large individual leaves (Cock et al., 1979), large root (Birador et al., 1978), numerous roots (IITA, 1976, Birador et al., 1978) and late branching (Lian & Cock 1979; Cock et al., 1979; IITA, 1980). Root yield also depends on the cultural practice adopted.

Root number, size, colour of outer cork and internal tissue vary greatly. Surface cork layers may be rough or smooth, white or cream and

light to dark brown in colour. The colour of the tuber surface is also a varietal characteristics, although brown is the predominant tuber surface colour. The cortex colour is usually white but may be tinged, pink or purple in colour. The internal tissue colour is usually white but may be yellow. A mature tuber (excluding the tail) may range in length from 5-100 cm and weigh from 0.5 to 2.0 kg depending on variety and growing conditions (Onwueme, 1978).

Tuberization involves the onset of secondary thickening in fibrous root. Thus the development of the tuber consists mainly of an increase in the diameter of a root. The actual number of roots, which eventually form tubers, depends on several factors, including cultivars; assimilate supply photoperiod and temperature (IITA, 1990). Generally, 4-10 tubers per plant may be produced (Purseglove, 1987; IITA, 1990). The number of roots that can potentially enlarge is fixed within three months of planting in varieties of 9-12 months duration (CIAT, 1973; Wholey & Cock, 1974; IITA, 1990). A critical percentage of assimilate supply is required for the initiation of tuberization. Therefore other factors, moisture stress, soil fertility status, soil aeration, soil temperature and radiation which affect assimilate supply also affect tuberization (IITA, 1990). Most varieties initiate tubers only under short day condition. Long days tend to encourage abundant shoot growth, delay tuber initiation and thus producing fewer tubers.

Wide variability in shoot morphology exists among cassava cultivars. There is great variation in stem colour, branching height and branching levels, plant height, angle of branching, the number of shoots per

plant and internodes length. Stem colour varies considerably ranging from whitish, grayish, light brown to dark brown (IITA, 1990). Branching height of mature plant may be very close to the ground or at a reasonable distance above the ground. Branching is influenced by several factors such as cultivar, time of planting, mutilocational fields, physical damage, environmental conditions including light, soil fertility, water stress and cool temperature and level of available photosynthate. The pattern of branching of the cassava plant varies with different cultivars (Jones, 1959). Cassava varies in height ranging from tall varieties (greater than 1.5 m) and short varieties (less than 1 m) (Okeke, 1979). The height of cassava varies not only genetically but also with environment condition such as altitude, temperature, insulation, soil fertility, lodging and if leaves are harvested or not (Nweke et al., 1994).

Variation in leaf characteristics is seen in leaf size, number of lobes, shape, width and length of lobes, colour of petiole, lamina, vein, and length of petiole (Purseglove, 1987). Petiole length varies from 5-30 cm, generally longer laminar lobes, which are usually 4-20 cm long and 1-6 cm wide (Onwueme, 1978). The amount of red pigment in the vein and petiole varies from the faintest traces to complete coloration. But in most varieties the central part of the petiole is coloured red or purple and the end are green apart from the extreme basal portion, which is red (Mason, 1956). Where the vein is reddish or purplish, the petiole is also reddish or purple in colour; otherwise the vein and petiole are greenish (Onwueme, 1978).

Association among characters in cassava

Association among various traits in cassava is essential in effecting selection. This relationship may reveal which of the phenotypic markers are indicators for certain agronomic traits. It would be of interest to know whether the improvement of one character will result in simultaneous changes in other characters. These associations could also identify characters that have little or no importance in a selection programme. The practical importance of selecting for a given trait as an indicator for a trait being sought however depends on the extent to which the improvement in the major traits being sought is facilitated by selecting for the indicators.

In cassava, the number of roots is significantly and positively correlated with the weight of root, top fresh weight, plant height and tuber size (Ntawuruhunga et al., 1995). Magoon et al. (1970) reported that yield was closely related to the number of storage roots even though the number of storage roots was inversely related to storage roots size. Several correlation studies in cassava have indicated that tuberous root yield is genetically related with number of tuberous roots per plant, tuberous root size, plant height and total number of branches (Magoon et al., 1970; Cock et al., 1979; Mahungu et al., 1987). Various reports have shown that pest and disease susceptibility is closely tied with low yield as yield depends on the level of pests and disease incidence (Akinlosutu, 1985; Odongo & Orone, 1992; Thresh et al., 1994). Disease resistance has been shown to be associated with pubescence of young leaves (Hahn & Keyser, 1985; Kanno et al., 1991).

Genetic Markers

Genetic improvement of cassava is to some extent limited by a poor knowledge of genetic diversity within the species. DNA markers have been useful in the evaluation of genetic diversity in crop species. According to Spooner et al. (2005), molecular markers present powerful new tools to reinvestigate hypotheses of hybridization. Marker assisted selection offers a great opportunity for imparting improved efficiency and effectiveness in selection of plant genotypes (Asante & Offei, 2003).

Due to the rapid developments in the field of molecular genetics, a variety of different techniques have emerged to analyze genetic variation during the last few decades (Whitkus et al., 1994; Karp et al., 1996; Parker et al., 1998; Schlötterer, 2004). Genetic markers can be classified into biochemical and molecular markers (Spooner et al., 2005). These genetic markers may differ with respect to important features, such as genomic abundance, level of polymorphism detected, locus specificity, reproducibility, technical requirements and financial investment. No marker is superior to all others for a wide range of applications. The most appropriate genetic marker will depend on the specific application, the presumed level of polymorphism, the presence of sufficient technical facilities and know-how, time constraints and financial limitations. The commonly used DNA markers in diversity analysis are RAPD, AFLPs, and SSRs (Raghu et al., 2007). The main marker technologies that have been widely applied during the last decades are briefly outlined below, together with their strengths and weaknesses.

Allozymes

Allozymes are allelic variants of enzymes encoded by structural genes. Enzymes are proteins consisting of amino acids, some of which are electrically charged. As a result, enzymes have a net electric charge, depending on the stretch of amino acids comprising the protein. When a mutation in the DNA results in an amino acid being replaced, the net electric charge of the protein may be modified, and the overall shape (conformation) of the molecule can change. Because changes in electric charge and conformation can affect the migration rate of proteins in an electric field, allelic variation can be detected by gel electrophoresis and subsequent enzyme-specific stains that contain substrate for the enzyme, cofactors and an oxidized salt. Therefore, allozyme variation is often also referred to as isozyme variation (Kephart, 1990; May, 1992).

The strength of allozymes is simplicity. Because allozyme analysis does not require DNA extraction or the availability of sequence information, primers or probes, they are quick and easy to use. Some species, however, can require considerable optimization of techniques for certain enzymes. Simple analytical procedures, allow some allozymes to be applied at relatively low costs, depending on the enzyme staining reagents used. Allozymes are codominant markers that have high reproducibility.

The main weakness of allozymes is their relatively low abundance and low level of polymorphism. Moreover, proteins with identical electrophoretic mobility (co-migration) may not be homologous for distantly related germplasm. In addition, their selective neutrality may be in

question (Berry & Kreitman, 1993; Hudson et al., 1994; Krieger & Ross, 2002). Allozymes are in fact phenotypic markers, and as such they may be affected by environmental conditions. For example, the banding profile obtained for a particular allozyme marker may change depending on the type of tissue used for the analysis (e.g. root or leaf). This is because a gene that is being expressed in one tissue might not be expressed in other tissues. On the contrary, molecular markers, because they are based on differences in the DNA sequence, are not environmentally influenced, which means that the same banding profiles can be expected at all times for the same genotype. With the advent of molecular biology techniques, DNA-based markers have replaced enzyme markers. Because of its plasticity, ubiquity and stability, DNA is the ideal molecule for such analysis

Restriction Fragment Length Polymorphism (RFLP)

RFLPs range 2–10 kb that has resulted from the digestion of genomic DNA with restriction enzymes. The DNA sequence variation affecting the absence or presence of recognition sites of restriction enzymes, and insertions and deletions within two adjacent restriction sites, form the basis of length polymorphisms.

RFLPs are generally found to be moderately polymorphic. In addition to their high genomic abundance and their random distribution, RFLPs have the advantages of showing codominant alleles and having high reproducibility.

However, RFLPs require laborious and technically demanding methodological procedures, and are highly expensive. In general, if research is conducted with poorly studied groups of wild species or poorly studied crops suitable probes may not yet be available, so considerable investments are needed for development. Moreover, large quantities (1–10 μg) of purified, high molecular weight DNA are required for each DNA digestion. Larger quantities are needed for species with larger genomes, and for the greater number of times needed to probe each blot. Another drawback is that RFLPs are not amenable to automation, hence, collaboration among research teams requires distribution of probes.

Random Amplified Polymorphic DNA (RAPD)

RAPDs are DNA fragments amplified by the PCR using short synthetic primers (generally 10 bp) of random sequence. These oligonucleotides are usually able to amplify fragments from 1–10 genomic sites simultaneously. Amplified fragments, usually within a range of 0.5–5 kb, are separated by agarose gel electrophoresis, and polymorphisms are detected, after ethidium bromide staining, as the presence or absence of bands of particular sizes. These polymorphisms are considered to be primarily due to variation in the primer annealing sites, but they can also be generated by length differences in the amplified sequence between primer annealing sites.

The main advantage of RAPDs is that they are quick and easy to assay. Because polymerase chain reaction (PCR) is involved, only low quantities of template DNA are required, usually 5–50 ng per reaction.

Since random primers are commercially available, no sequence data for primer construction are needed. Moreover, RAPDs have a very high genomic abundance and are randomly distributed throughout the genome.

The main drawback of RAPDs is their low reproducibility (Schierwater & Ender, 1993), and hence highly standardized experimental procedures are needed because of their sensitivity to the reaction conditions. RAPD analyses generally require purified, high molecular weight DNA, and precautions are needed to avoid contamination of DNA samples because short random primers are used that are able to amplify DNA fragments in a variety of organisms. Altogether, the inherent problems of reproducibility make RAPDs unsuitable markers for transference or comparison of results among research teams working with a similar species and subject. Another disadvantage is that RAPD markers are not locus-specific, band profiles cannot be interpreted in terms of loci and alleles (dominance of markers), and similar sized fragments may not be homologous. Therefore, RAPD markers are not used routinely anymore because of their inconsistency from laboratory to laboratory.

Amplified Fragment Length Polymorphism (AFLP)

AFLPs are DNA fragments (80–500 bp) obtained from digestion with restriction enzymes, followed by ligation of oligonucleotide adapters to the digestion products and selective amplification by PCR. AFLPs, therefore, involve both RFLP and PCR. The PCR primers consist of a core sequence and a restriction enzyme specific sequence and 1 to 5 selective nucleotides. The higher the number of selective nucleotides, the lower the

number of bands obtained per profile. The AFLP banding profiles are the result of variations in the restriction sites or in the intervening region. The AFLP technique simultaneously generates fragments from many genomic sites (usually 50 to 100 fragments per reaction) that are separated by polyacrylamide gel electrophoresis and that are generally scored as dominant markers.

The strengths of AFLPs lie in their high genomic abundance, considerable reproducibility, the generation of many informative bands per reaction, their wide range of applications, and the fact that no sequence data for primer construction are required. AFLPs may not be totally randomly distributed around the genome as clustering in certain genomic regions, such as centromeres, has been reported for some crops (Alonso-Blanco et al., 1998; Young et al., 1999; Saal & Wricke, 1999). AFLPs can be analyzed on automatic sequencers, but software problems concerning the scoring of AFLPs are encountered on some systems.

The disadvantages include the need for purified, high molecular weight DNA, the dominance of alleles, and the possible non-homology of comigrating fragments belonging to different loci. In addition, due to the high number and different intensity of bands per primer combination, there is the need to adopt certain strict but subjectively determined criteria for acceptance of bands in the analysis.

Microsatellites

Microsatellites represent tandem repeats, but their repeat motifs are shorter (1 - 6 bp). If nucleotide sequences in the flanking regions of the

microsatellite are known, specific primers (generally 20–25 bp) can be designed to amplify the microsatellite by PCR. Microsatellites and their flanking sequences can be identified by constructing a small-insert genomic library, screening the library with a synthetically labelled oligonucleotide repeat and sequencing the positive clones. Alternatively, microsatellites may be identified by screening sequence databases for microsatellite sequence motifs from which adjacent primers may then be designed. In addition, primers may be used that have already been designed for closely related species (Matsuoka et al., 2002).

The strengths of microsatellites include the codominance of alleles, their high genomic abundance in eukaryotes and their random distribution throughout the genome, with preferential association in low-copy regions (Morgante et al., 2002). Because the technique is PCR-based, only low quantities of template DNA (10–100 ng per reaction) are required. Due to the use of long PCR primers, the reproducibility of microsatellites is high and analyses do not require high quality DNA. Among the recently developed molecular markers, Simple sequence repeat (SSR) markers are being considered as the markers of choice for many types of investigations, including marker-assisted selection and finger-printing of germplasm collections, as it detects the variation in allele frequency at many unlinked loci and provides high information content (Kong et al., 2000). SSR markers are particularly attractive to study because they are abundant in plants, codominant, have high level of polymorphism, and are adaptable to automation (Hearne et al., 1992; Morgante & Olivieri 1993; Queller et al., 1993; Jarne & Lagoda 1996; Donimi et al., 1998). According to Raghu et

al. (2007) there are more than 500 SSR markers available in cassava, which will help to have genetic tags for various phenotypes in cassava. Although microsatellite analysis is, in principle, a single-locus technique, multiple microsatellites may be multiplexed during PCR or gel electrophoresis if the size ranges of the alleles of different loci do not overlap (Ghislain et al., 2004). This decreases significantly the analytical costs.

One of the main drawbacks of microsatellites is the high development costs, particularly if adequate primer sequences for the species of interest are unavailable, thus making them difficult to apply to unstudied groups. However, once developed, analysis of them is both easy and inexpensive (Kong et al., 2000). A very common observation in microsatellite analysis is the appearance of stutter bands that occur by DNA slippage during PCR amplification. These can complicate the interpretation of the band profiles because size determination of the fragments is more difficult (Sponner et al., 2005).

The use of genetic markers in cassava improvement

A prerequisite to any genetic improvement programme for cassava is the knowledge of the extent of genetic variations present within the *Manihot* species. These are important and have been applied using diverse number of molecular markers. The following genetic studies have been carried out in cassava; the identification of close relatives of cassava with which good hybrids could be produced and from where introgression of desirable traits into cassava is possible (RAPD, Tonukari et al., 1997; SSR and RAPD, Carvalho & Schaal, 2001), characterization of germplasm to

develop large number of new varieties with diverse agronomic traits (AFLP, Angel et al., 1992; Elias et al., 2000; AFLP and RAPD Colombo et al., 2000; RAPD and RFLP, SSR, Fregene et al., 2001; RAPD, Asante & Offei, 2003), for identification of duplicates and elimination of dormancy (RAPD, Ugorji, 1998; AFLP and SSRs, Chavarriaga-Aguirre et al., 1999; AFLP, Fregene et al., 2000), and identification of representative set for development core collection (RAPD, Colombo et al., 2000; RAPD and SSR, Carvalho & Schaal, 2001). Molecular markers have also been used in cassava in disease diagnosis and identification of source of resistance to diseases and pests and other agronomic traits (AFLP Fregene et al., 2000; SSRs, Fregene et al., 2001; AFLP and SSR Lokko et al., 2005; Okogbenin et al., 2006). Again DNA-based molecular markers such as RAPDs, nuclear RFLPs and microsatellites have been used to develop the cassava molecular genetic map (Fregene et al., 1997). Similarly, RAPDs, AFLPs, cpDNA and cDNA RFLPs have been used to assess the genetic variability of small sets of cassava germplasm and to establish relationships between cassava and its wild relatives (Beeching et al., 1993; Bertram, 1993; Fregene et al., 1994; Marmey et al., 1994; Roa et al., 1997).

RAPD markers are not used routinely anymore because of their inconsistency from laboratory to laboratory, as mentioned earlier. Simple Sequence Repeat (SSR) markers have been confirmed to be the most informative and appropriate for cassava (Mba et al., 2000). Perera et al. (2000) also support SSR markers as the most informative for plants. Valuable attributes of all SSR markers are codominance (many alleles are found among closely related individuals), technical simplicity, sensitivity,

analytical simplicity (data are unambiguously scored, and highly reproducible) and are high in abundance (markers are uniformly dispersed throughout genome) as frequently as every 10 kb and therefore are ideal tools for many genetic applications.

Various research studies have indicated that the number of DNA markers used for genetic studies in plants varies with the total number of accessions assessed (Thottapilly et al., 1996). Hence, the larger the number of plant accessions assessed the larger the number of DNA markers utilized. Generally, large numbers of SSR primers are being utilized for genetic diversity and genotype identification studies that ranged from 6 for almond (Joobeur et al., 2000) to 136 for sunflower (Yu et al., 2000). Ahmad (2002) used 43 SSR primers on 13 genotypes of wheat and proposed that a collection of polymorphic SSR primers could be used for genotype identification, genetic diversity, and map linkage studies.

Effect of mosaic virus disease on chlorophyll content of cassava

Plant pigments mainly include chlorophyll, carotenoids, and flavonoids. Chlorophyll is the most abundant pigment found in plants, some algae, and some bacteria. It is this that gives them their green colour and that absorbs the light necessary for photosynthesis. Chlorophyll absorbs mainly violet-blue and orange-red light. The great abundance of chlorophyll in leaves and its occasional presence in other plant tissues, such as stems, causes these plant parts to appear green. In some leaves, chlorophyll is masked by other pigments (Cosby & Raven, 2007).

Chlorophyll is a large molecule composed mostly of carbon and hydrogen. At the centre of the molecule is a single atom of magnesium surrounded by a nitrogen-containing group of atoms called a porphyrin ring. There are two main forms, chlorophyll *a* and chlorophyll *b*, normally present in the ratio to about 3:1.

Due to the green colour of chlorophyll, it has many uses as dyes and pigments. It is used in colouring soaps, oils, waxes and confectionary. Chlorophyll's most important use, however, is in nature, in photosynthesis. It is capable of channelling the energy of sunlight into chemical energy through the process of photosynthesis. In this process the energy absorbed by chlorophyll transforms carbon dioxide and water into carbohydrates and oxygen.

Chlorophyll determination is usually carried out on fresh material, because drying with heat tends to degrade the pigment resulting in colour change (Stewart et al., 1974). If fresh material has to be stored, deep-freezing at -20°C to -30°C is most appropriate Goodwin, (1963). However, if drying is unavoidable freeze-drying is probably the best method (Stewart et al., 1974).

The intense green colour of chlorophyll is due to its strong absorbencies in the red and blue regions of the spectrum, shown in Figure 1. Because of these absorbencies the light it reflects and transmits appears green.

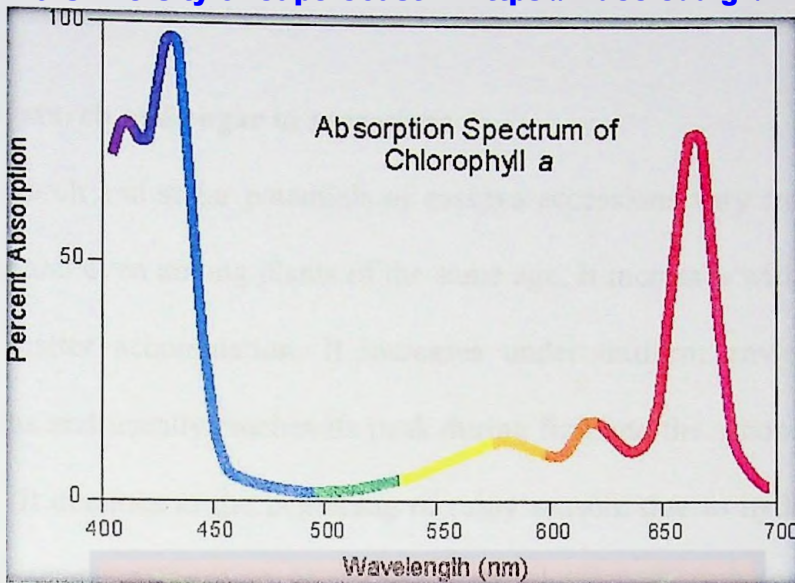


Figure 1: The UV/visible adsorption spectrum for chlorophyll

Leaf chlorophyll content influences photosynthesis and light absorption and is often indirectly assorted with growth and yield of cassava (Ekanayake et al., 1996; Oyetunji et al., 1998). Decreases in photosynthetic activity are often paralleled by a reduction in leaf chlorophyll content (Ekanayake et al., 1996). Ekanayake et al. (1996) have noted that low moisture condition, high air and soil temperatures at midday and very low temperatures at night, during the harmattan period; negatively affect chlorophyll fluorescence activities, which contribute to reduction in growth rate of cassava during the dry season in the upland.

Cassava mosaic virus has variable effects on the chlorophyll content of cassava. Ayanru and Sharma (1982), have found that the ratio of chlorophyll *a* and chlorophyll *b* were similar in diseased and healthy leaves. Yan-Ping et al. (2004) have found a significant reduction in chlorophyll content in viral-infected plants.

Starch

Level of starch and sugar in accessions

Starch and sugar potentials of cassava accessions vary from plants to plants and even among plants of the same age. It increases with increase in dry matter accumulation. It increases under uniform environmental conditions and usually reaches its peak during first and the second bulking phases. It declines at the beginning of rainy season, due to hydrolysis of starch to sugar (Henry et al., 1998). Starch and sugar production is increased when the soil has higher potassium level because potassium helps in formation of more starch vacuoles (Addo-Quaye et al., 1993).

Sugar, starch and protein contents of cassava flour obtained from different accessions at IITA ranged from 2.4-5.1%, 61.8 – 66.4% and 29.3 – 36.6%, respectively (IITA, 2001).

Cassava root loose starch when they are left in the ground after maturity (IITA, 2001). Since root do not accumulate starch after maturity but sugar. The presence of green leaves of cassava at final harvest indicates a continuing capacity for photosynthesis, which leads to accumulation of sugar with time. As a result high levels of total sugar are concentrated in roots and leaves after maturity, which are not converted to starch. Since sugar is soluble, it is easily eliminated through food processing methods. Therefore, the practice of harvesting early maturing varieties about nine months after planting needs to be reviewed, because only starch component is nutritionally relevant to human (Ketiku & Oyenuga, 1972)

Properties of starch

Starch is a polysaccharide composed of a long chain of glucose molecules. Starch is the storage form of carbohydrates in plants. The starch that is produced by the plant often occurs as grains within the cell. The grains are built up as concentric layers of starch molecules that are found within membrane-bound organelles called amyloplasts (leucoplast) in the cytoplasm. Pure isolated starch is a white, amorphous, relatively tasteless solid which possesses no odour, and which is insoluble in cold water. Starch granules are characterized by a birefringence, that is, the ability to refract polarized light (Rickard et al., 1991). The birefringence indicates that the granule has a high degree of molecular orientation (Lineback, 1984).

Chemically, starches are polysaccharides that consist of repeating glucose units. Starch molecules have one of two molecular structures: a linear structure, known as amylose; and a branched structure, known as amylopectin. Amylose and amylopectin associate through hydrogen bonding and arrange themselves radially in layers to form granules. Starches from different sources vary from one another in the following ways: granule size and shape, amylose:amylopectin ratio and structure of amylose and amylopectin which may affect performance.

Chemical Structure of Starch

Chemically, starch is a homopolysaccharide mainly composed of amylose and amylopectin units. The functionality of starch depends on the content of these polymers.

Amylose

Amylose is a fraction of starch which is composed of repeating glucose molecules linked with α -glucose molecules linked together by 1,4-linkages (α -1,4-linkages) (Manners, 1968; Kaufman, 1989). Figure 2 shows the chemical structure of amylose. Molecular weight determinations indicate that the amylose has a degree of polymerization of up to DP 6000 and a molecular mass of around 105 g mol^{-1} (Sajilata et al., 2006). The amylose content in native starch can range anywhere from 0 to almost 50%. Amylose forms stiff and generally irreversible gels that will only reverse upon heating to autoclave temperatures (110 - 160 °C). The tightly bonded structures and intimate associations promote the stiffness and irreversibility of the gels (Zobel, 1988).

There may be a slight degree of branching in the amylose molecule (a very few α -1, 6 branches and linked phosphate groups), but these have little influence on the molecule's behaviour (Buléon et al., 1998). The molecule is coiled in the shape of a flexible helix with a period of six to seven units. The interior of the helix contains predominantly hydrogen atoms and is lipophylic, while the hydroxyl groups are positioned on the exterior of the coil (Whistler et al., 1984)

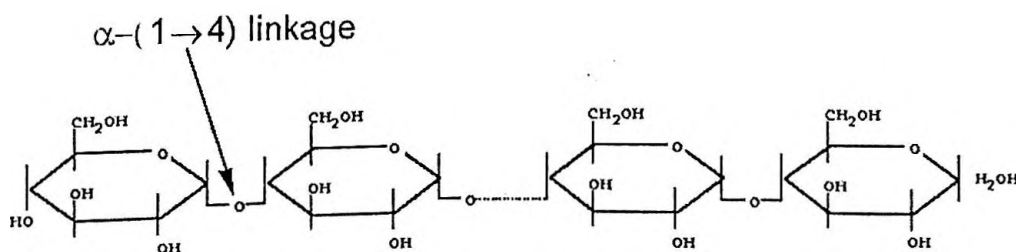


Figure 2: Structure of Amylose
Adapted from Nowjee (2004)

There is enough space for an iodine molecule inside each flexible coil. This characteristic forms the basis for the starch test (Freeland-Graves & Peckham, 1987). Thus, if iodine is added to a solution containing starch, the iodine is inserted within the coil and makes it rigid. This transformation colours the starch mixture blue if the helix (or glucose chain) is long or reddish purple if the helix length is short.

Amylopectin

Amylopectin is the second fraction that is found within starch. This polysaccharide is a polymer with glucose molecules linked together with α -D (1-4) and α -D (1-6) linkages (Figure 3). Amylopectin is highly branched and has a degree of polymerization (DP) of 2 million and a molecular weight of around 109 g/mol making it one of the largest molecules found in nature. The structure of amylopectin is characterized by a central chain of glucose molecules held together with α -D (1-4) linkages with branches at every 20-25 glucose units that come off of the main chain with α -D (1-6) linkages (Kaufman, et al., 1989; Sajilata et al., 2006). The amount of amylopectin present in a starch can be as low as 50 % and as high as 100 %. Starches with 100% amylopectin are known as waxy starches.

Amylopectin forms soft, reversible gels; these polymers do not complex readily. The softer gels are due to the highly branched nature of the polymer making interactions less favourable and fewer in numbers. The temperature required to reverse an amylopectin gel can range anywhere

from room temperature to 90 °C, depending on the degree of polymerization and the number of branches of the particular amylopectin (Zobel, 1988). Table 2 shows the properties of amylose and amylopectin.

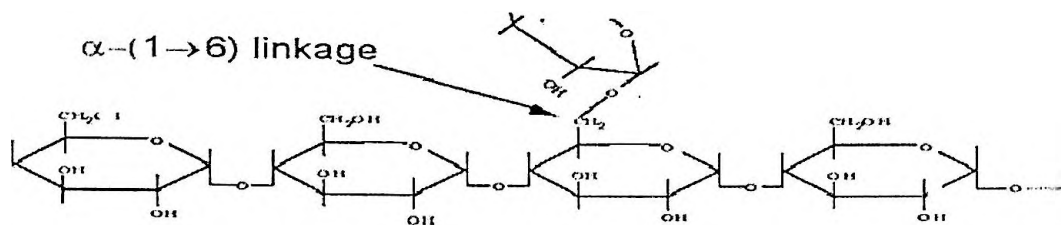


Figure 3: Structure of Amylopectin

Adapted from Nowjee (2004)

Table 2: Properties of Amylose and Amylopectin

Properties	Amylose	Amylopectin
General structure	Essentially linear	Branched
Colour with iodine	Dark blue	Purple
Maximum of iodine complex	Approx. 650nm	Approx. 540nm
Iodine affinity	19 – 20%	<1%
Average chain length	100 – 10,000	20 – 30
Degree of polymerization (DP)	100 – 10,000	10,000 – 100,000
Solubility in water	Variable	Retrogrades
Solubility in aqueous solution	Retrograde	Stable
Conversion to maltose by (Crystalline B-amylose)	Approx. 70 %	Approx. 55 %

(Shannon and Garwood, 1984)

Gallant et al. (1997) have postulated that amylopectin is arranged in the granule as clusters of radially oriented chains organized in super helical and semi-crystalline blocks. The proposed model has emerged mainly from chain length distribution analysis of debranched amylopectin (Hizukuri, 1986), electron microscopy (Oostergetel & Van Bruggen, 1993), polarized light microscopy (French, 1972), electron diffraction microscopy

and fibre X-ray crystallography (Imberty et al., 1988; Imberty & Perez, 1988; Imberty et al., 1991).

The relative proportion of amylose and amylopectin in starches are responsible for the differences in cooking characteristics of the different types of starches. According to Hegenbart (2009), longer amylose molecules tend to make a product's texture stringy because of the way they associate. The molecular weight of the amylose also affects the elasticity of a gel. Starches containing a higher percentage of amylopectin have a higher peak viscosity and paste stability; this means that the starch will produce a thicker paste which will be less likely to break down during cooking (Bainbridge et al., 1996). Amylose becomes cloudy when heated with water and is capable of forming a gel. Amylopectin, however, remains clear when heated with water and does not set a liquid or gel.

Amylose amylopectin ratio

Amylose:amylopectin ratio is an important aspect of starch that influences its property and function. The amylose:amylopectin ratio determines not just the basic texture, but the nature of that texture, as well. For instance, waxy corn and common corn starches both have the same granule size, but waxy corn will swell to a greater degree and each will gelatinize at different temperatures. This is largely due to their differing amylose: amylopectin composition.

Amylose molecules, because of their linearity, line up more readily and have more extensive hydrogen bonding. Consequently, it requires more energy to break these bonds and gelatinize the starch (Hegenbart, 2009).

Generally, the higher the amylose, the higher the gelatinization temperature. This is most noticeable in the two high-amylose corn starches which require such high temperatures for gelatinization that they must be cooked under pressure. The amylose:amylopectin ratio also determines the sort of texture the gelatinized starch will build.

According to Hegenbart (2009), amylose gives you the gel strength and the amylopectin gives you high viscosity. So the high-amylose starches will give you gelling properties and the waxy starches will give you high viscosity. The linear structure of amylose also contributes to gel strength. In solution, the linear amylose molecules can more easily align themselves with one another and associate through hydrogen bonding to form gels. The branched amylopectin molecules cannot align as easily and, thus, give weaker hydrogen bonding and gel strength.

Viscosity, on the other hand, is purely a function of molecular weight. The branched structure of amylopectin with all its attached chains yields a much larger molecule than amylose. Consequently, amylopectin is better at building viscosity than amylose.

Rheological Characteristics of Cassava Starch

According to Dengate et al. (1984) most of the functional attributes of starch can be related to the temperature-dependent interactions of starch with water, the process known as pasting, gelatinization and retrogradation. As starch is heated in the presence of water, granule swell and imbibe water, hydrogen bonds are disrupted with eventual irreversibly loss of crystallite structure (Atwell et al., 1988).

Swelling Power

Swelling power provides evidence of non-covalent bonding between starch molecules; it is affected by factors such as amylose-amylopectin ratio and chain length. Molecular weight distribution, degree of branching and conformation determine the degree of swelling and solubility (Rickard et al., 1991). They reported that the swelling power of cassava varies considerably from 42-71 g/g and this is within those of potato and some cereal starches. They also observed that during the growth period, starches of some varieties maintain their swelling volume within small ranges while others expressed wide variation. This means that the environment has an influence on varieties. Sanni et al., (2005) reported that the swelling index of granules reflect the extent of associative forces within the granules, therefore the higher the swelling index, the lower the associative forces.

Asaoka et al. (1992) have determined swelling volume of four varieties of cassava harvested during two seasons. It was found that the swelling power was higher for samples harvested in the month November compared to those harvested in August. High amylose content and the presence of stronger or higher number of intermolecular bonds can reduce swelling power (Delpeuch & Favier, 1980). The formation of liquid-starch complex and the presence of naturally occurring carbohydrates and non-carbohydrates can also affect the swelling volumes (Eliassan & Gudmundsson, 1996).

Solubility

Solubility of starch depends on a number of factors such as inter-associative forces swelling power and the presence of other components. Cassava starch has a higher solubility than other tuber crop starches and the higher solubility can be attributed partly to the high swelling cassava starch undergoes during gelatinization (Moorthy, 2001).

The solubility of starch from different cassava varieties varied from 17.2-27.6 per cent. However, no direct correlation was found between swelling and solubility (Moorthy, 2001). In previous presentations, he observed that, among a few non-aqueous solvents studied for solubility of starch, maximum solubility was obtained in DMSO and formalin, while in glycerol it was moderate. He also observed that starch was insoluble in anisole and methylcellosolve; this indicates that starch is more soluble in polar solvents or solvents with affinity towards water.

The solubility of starch extracted from seven sweet potato collections from Peru indicated that solubility increased with temperature and reached nearly 10 per cent, while that for commercial starches, was 28 per cent (Garcia & Walter, 1998). They reported that selection identity did not have noticeable effect, but location had significant influence at above 60 °C.

Swelling behaviour has been found to be dependent on the nature and strength of the structural arrangement within the starch granules, and hence, dependent on the strength and nature of the associative forces within the starch granules (Leach et al., 1959). Moorthy and Ramanujam (1986)

reported that swelling power and solubility of cassava starch are dependent on varietal differences, environmental factors and the age of the crop.

Starch Gelatinization

Starch gelatinization is a process that breaks down the intermolecular bonds of starch molecules in the presence of water and heat, allowing the hydrogen bonding sites (the hydroxyl hydrogen and oxygen) to engage more water. During the cooking of starch mixtures, several changes take place that are significant in the preparation of typical starch products. When starch granules are added to cold water, a small amount of water is absorbed causing a reversible swelling. A temporary suspension in which the starch granules do not dissolve is also formed. The starch tends to settle out of the mixture as soon as the mixture is allowed to stand (Freeland-Graves & Peckham, 1987).

When the starch mixture is heated, the water begins to penetrate the starch granules in quantity, causing them to swell and lose their birefringence. Penetration of water increases randomness in the general structure and decreases the number and size of crystalline regions. Crystalline regions do not allow water entry. Heat causes such regions to be diffused, so that the chains begin to separate into an amorphous form. Swelling is reversible up to the point at which the molecular structure within the granules is disrupted and birefringence is lost. Over a relatively narrow temperature range, all the granules swell irreversibly and are said to have undergone gelatinization. Continued heating of the gelatinized starch grains (*pasting*) causes the starch granules to swell enormously and soften,

forming a viscous paste. If the paste is fluid, it is called a sol; if it is solid, it is called a gel.

The primary event that occurs when starch is gelatinized in an aqueous medium is granule swelling. As the temperature of an aqueous suspension of starch is raised above the gelatinization or pasting range, hydrogen bonds continue to be disrupted, water molecules become attached to the liberated hydrogen groups and the granules continue to swell. As a direct result of granule swelling, there is a parallel increase in starch solubility, paste clarity and paste viscosity (Knight, 1969; Mat-Hashim et al., 1992). Also the additional increase in the viscosity of the starch paste with further heating is believed to be the result of starch being exuded out of the starch grain into the surrounding medium. The starch molecules trap the free water and inhibit its free flow.

Perhaps the most important variable characteristics of different starches when observed are the ways the starches form paste when heated with water. The differences are evident in a number of ways: in the temperatures at which the granules start to swell; the way the viscosity increases as the temperature increases and more granules become hydrated; the way the viscosity increases as the paste cools; and the degree to which the paste breaks down under the effect of shearing actions (Jones, 1983).

The process of gelatinization and pasting vary with the type of starch and size of the starch granule. Generally, starches with large granules swell at lower temperatures than those with smaller granules. For example, potato, waxy corn, and tapioca starch thicken at much lower temperatures than do regular corn and wheat starch. Continued heating of the starch

mixture after it has achieved its peak viscosity will decrease the thickness of the starch paste. The ability of starch to swell and produce a viscous paste when heated in water (or treated with certain chemicals) is its most important practical use in the food industry since they affect the texture and digestibility of starchy foods.

Gelatinization and Pasting temperature

Gelatinization of starch takes place over a definite range of temperature known as gelatinization temperature. The gelatinization temperature is indicative of the temperature at which part of amylose separates from amylopectin and leaches out of the starch granules during heating. The pasting (or peak gelatinization) temperature on the other hand, is the temperature at which irreversible swelling of the starch granules occur leading to peak viscosity. Pasting temperature is always higher than gelatinization temperature (Moorthy, 2001).

Moorthy (1994) studied the gelatinization temperatures of starch of seven cassava varieties and found out that two varieties gelatinized earlier with gelatinization temperature range of 12°C. No relationship between granule size and gelatinization temperature was observed. Pasting temperatures were also determined using a viscograph and all the values obtained were similar except one variety that showed a lower pasting temperature.

According to Bainbridge et al. (1996), an increase in viscosity indicates the tendency of the starch samples to retrograde or associate.

They reported that generally, starch with high pasting temperature and high peak viscosity has weak associative forces.

Pasting temperatures of starch from four varieties of cassava commonly cultivated in Ghana were found to range from 64 to 67 °C (Boakye et al., 2001). Working on seven varieties of a related root and tuber crop, sweet potato, Oduro et al. (2000) observed that the pasting temperatures were relatively high and varied between 72 and 73.3 °C.

According to Bainbridge et al. (1996) starches with lower pasting temperatures are generally considered to be easier to cook. However, lower pasting temperatures are also associated with low paste stability, which is usually considered to be an undesirable property. Low pasting temperature and low paste stability indicate that fewer associative force and cross-links are present within the starch granule.

Generally, the higher the amylose, the higher the gelatinization temperature. Amylose molecules, because of their linearity, line up more readily and have more extensive hydrogen bonding and consequently require more energy to break these bonds and gelatinize the starch (Hegenbart, 2009).

Paste viscosity

An important property of starch is that it provides a viscous paste (thickened starch mixture) when heated in presence of water. It is this viscosity which accounts for the use of starch in textile, paper, adhesive and food industries. Cassava is well known for high viscosity of its paste.

When starch of different varieties of cassava was studied using a Brabender Viscograph three peak patterns were generally observed (Moorthy, 1994). They were: -

- (i) Single stage gelatinization with high peak viscosity and high viscosity breakdown.
- (ii) Two-stage gelatinization with high peak viscosity and breakdown.
- (iii) Broad two-stage gelatinization with medium viscosity and medium breakdown.

Moorthy (1994) in another study involving five varieties of cassava having different cooking quality observed that the starch of one variety had a medium peak viscosity, low viscosity breakdown but high setback viscosity. Another variety had slightly lower peak viscosity and setback viscosity. A third variety, on the other hand, had a very high peak viscosity which tinned down considerably on heating and the setback viscosity was low. The results seemed to indicate some relationship between cooking quality and starch rheology since the variety with medium peak viscosity and high setback viscosity reasonably had good cooking quality compared to the variety with high peak viscosity and low setback viscosity which had poor culinary quality.

Viscosity analysis

The peak viscosity is the highest viscosity reached during the heating phase of the Brabender Visco-Amylograph. At this point, there is a majority of granules that are fully swollen but intact. For any particular type of starch, the more granules that are available to be hydrated the

higher the peak viscosity will be. During the high temperature hold phase at 95 °C, the starch granules begin to breakdown and solubilisation continues resulting in a drop in viscosity and a trough viscosity is recorded. The peak viscosity value and viscosity at 95 °C are measures of the ability of the starch to form a paste on cooking. The higher the value the thicker the paste will be. Jones (1983) and Kim et al., (1995) have noted that a high viscosity is desirable for industrial use, for which a high thickening power is required. The difference between the peak and trough viscosities is termed the “breakdown”. The rate of decrease in viscosity depends on the temperature and the nature of the material itself.

During the cooling phase, the solubilised starch molecules begin to reassociate and the viscosity begins to increase again towards the cold paste or final viscosity. In sufficient concentration, this usually causes the formation of a gel. This second rise, representing the difference between the paste and hot paste viscosities is known as the setback (retrogradation).

The main component that is said to retro grade is the amylose. Its linear structure can reassociate tightly forming a harder, firmer gel (Thomas & Atwell, 1998). Ring (1993) reported that highly branched chains of the amylopectin molecule project out too much and interfere with bonding to other molecules. Starches that contain higher amylopectin to amylose ratios tend to retrograde much slower than starches that have a high percentage of amylose; this is due to the highly branched nature of amylopectin that takes longer periods of time to reassociate in a tight manner (Moorthy, 2001). Retrogradation can have a major effect on the overall quality and shelf-life stability of food products. Retrogradation is often an undesirable side

effect of starch gels; this process when found in bread and other bakery products is known as staling and negatively affects the product (Katayama et al., 2002). A retrograded starch often exhibits the B-type crystalline pattern even when no amylose is present (i.e. waxy starch) (Annison and Topping, 1994).

When a cooled starch gel that has been standing for a while is cut, there is leakage of liquid from the gel. This leakage or separation of fluid from a gel is called syneresis or weeping (Freeland-Graves & Peckham, 1987).

A low setback value shows that the starch gives a non-cohesive paste which is useful in many industrial applications (Kim et al., 1995). A high setback value is useful if the starch is to be used in domestic products such as fufu, which require a high viscosity and paste stability at low temperature (Oduro et al., 2000).

In addition to the peak, stability of viscosity is also a very important factor which decides the applicability of starch in food and industry. Paste stability is determined by subtracting the viscosity value after 15 minutes at 95°C from the value for paste viscosity at 95°C (Oduro et al., 2000). The paste stability at 95°C measures the tendency of the paste to break down during cooking. High paste stability is frequently a requirement for industrial uses of starch.

A starch with low paste stability has very weak cross-linking within the granules and requires less heating. In this respect cassava starch is inferior to maize starch because its viscosity is rapidly reduced on heating under shear showing that the strength of associative forces is not very high.

This leads to a long and cohesive texture for its paste, which is not desirable in food and textile applications (Moorthy, 1994).

Boakye et al. (2001) have demonstrated that pasting behaviour of starch from four varieties of cassava, namely: 'Akosua Tumtum', 'Ankra', 'Abosome Nsia' and 'Adwoa Smart' showed significant variations ($p < 0.05$) in peak viscosity and viscosity at 95°C. Values recorded for peak viscosity and viscosity at 95°C ranged from 320 to 585 BU. The cold paste viscosities were very high for all the samples indicating the tendency of the starch samples to associate or retrograde on cooling.

In another work, Adomako (2007) studied the pasting characteristics of starch from seven accessions and one variety of cassava observed significant ($p < 0.05$) variations in peak viscosity and viscosity at 95°C. He reported peak viscosity values that ranged between 830.0 and 988.67BU, and variation of 1192.33 to 1398.67 BU at Legon and Bunso respectively. In another work, Oduro et al. (2000) studied the pasting characteristics of starch from seven new varieties of sweet potato and observed that the peak viscosity and viscosity at 95°C ranged from 480 BU for variety 'Dugbadza' to 600 BU for 'Sauti'. After the onset of pasting, the viscosity of all the samples increased rapidly, but the viscosity at 95 °C and after the first holding periods was lower than the peak viscosity, reflecting the strength of the starch pastes. Based on other physiochemical properties and the pasting characteristics of the seven sweet potato varieties, they concluded that variety L/Red will be suitable for domestic applications while 86/0250 will be better for many industrial purposes.

Sweet Potato Starch

Sweet potato starch is a very important food product material around the world with an emphasis of use in Asian countries (Moorthy, 2001). The usefulness of sweet potato starch ranges from one variety to another. The starch's use is determined by several factors including the amylose/amylopectin ratio, granule size, and the structure of the starch (Katayama et al., 2002). The sweet potato starch granules vary in shape from polygonal, round to oval with diameters ranging from 2 to 25 μm (Moorthy, 2001). The average sweet potato starch granule has an amylose content of around 18 % (Tsou & Hong, 1992). Sweet potato starch has been characterized as having various x-ray diffraction patterns ranging from A, C, and an intermediate between the A and C types. Also, the absolute crystallinity of this starch is reportedly around 38 %. Depending on variety, sweet potato starch has a known range of amylose content of 8.5-38 %, a gelatinization temperature of 63-79 $^{\circ}\text{C}$, and a pasting temperature of 58.5 to 90 $^{\circ}\text{C}$ (Moorthy, 2001).

Much research has been performed on the sweet potato across the world including much research on gelatinization, pasting, and retrogradation, and also on the various factors that can influence the properties of the sweet potato starch. Kaur et al. (2006) studied the gelatinization patterns of sweet potato starch and found that starches with high amylose content had a higher gelatinization temperature and a lower enthalpy than starches with lower amylose contents. The researchers reasoned that the correlation between amylose content with gelatinization temperature and enthalpy suggests that there must be a higher percentage of

amorphous regions rather than crystalline regions within the amylose. This lack of crystalline regions raises the gelatinization temperature (Kaur et al., 2006). The crystallinity of a starch granule imparts stability to the system that can be disturbed only through higher temperatures. Also, a correlation between the harvesting of sweet potatoes, early to late during the sweet potato growth period, and the enthalpy needed to gelatinize the starch has been noticed.

Research found that the earlier a sweet potato was harvested in its growing season, the lower the enthalpy will be to produce a gelatinized sample (Moorthy, 2001). Jangchud et al. (2003) found that the peak temperatures of pasting varied between sweet potato starches because of the variety of starch granule sizes that exist. Starches with larger granules were correlated with lower pasting temperatures, but also with an increase in the amount of swelling observed (Jangchud et al., 2003). Collado et al. (1999) found that the pasting viscosity and amylose content of sweet potatoes was negatively correlated. Moorthy (2001) reported that sweet potato starches having lower amylose contents or those starches with smaller amylopectin molecules retrograded slower than those starches having high amylose content.

Modification of starches

According to (Jensen, 2009) native starch swells quickly and loses its viscosity almost as quickly again during continued heating and forms a viscous coherent gel. This quality is perfect in some applications, but undesirable in other applications. Native starch is a good texture stabilizer

and regulator in food systems (Cousidine, 1982), but limitations such as low shear resistance, thermal resistance, thermal decomposition and high tendency towards retrogradation limit its use in some industrial food applications. Positive attributes of starches can be greatly improved and/or negative characteristics diminished by slight and relatively simple modifications.

According to (Jensen, 2009) when you modify the starch you adjust the properties of the starch to fit specific applications and to resist some of the undesirable characteristics of native starch. For modification purposes, native starch of the specification summarized in Table 3 is used. Starches are modified for a number of reasons. Starches may be modified to increase their stability against excessive heat, acid, shear, time and cooling or freezing; to change their texture; to decrease the viscosity, or to lengthen or shorten gelatinization time.

Table 3: Standard specification of native cassava starch for modification purpose

Property	Specification
Moisture content (% maximum)	13 %
Ash (% maximum)	0.2 %
Whiteness (Kett scale, minimum)	90
Viscosity (Barbender Unit, minimum)	600
Sulfur dioxide content (ppm, maximum)	100
Residue (ppm, maximum)	300

Adapted from Sriroth et al., 2002

Starch modification, which involves the alteration of the physical and chemical characteristics of the native starch to improve its functional characteristics, can be used to tailor starch to specific food applications (Hermansson & Svegmarm, 1996). The methods that are available for the

modification of starch can be non-degradative, using physical treatments, incorporation of chemicals and chemical treatments.

Starch modification is generally achieved through physical, chemical and enzyme process. Physical modified starches involves the treatment of cassava starch by physical means such as shear force, blending and thermal treatment. Chemical modified starch is prepared by chemical reaction. The most popular are oxidized starch and acid-modified starch. The chemical treatments are based on the availability of a large number of hydroxyl groups in the starch molecules, which can be made to react in many ways with various reagents.

Physical modification: starch blends

Physically modified starches involves the treatment of cassava starch by physical means such as shear force, blending and thermal treatment.

Starch is used as an additive in many food products to achieve certain characteristic properties, for example, water holding capacity, and specific rheological properties. In many applications the properties of a native starch are not optimal, and therefore starch is often chemically modified in order to improve its performance (Jacobs & Delcour 1998). Nowadays market's tendencies press the producers towards more natural food components, avoiding as much as possible the chemical treatments. It is therefore of interest to find new ways to improve the properties of native starches without using chemical modification. One possibility that is currently being investigated is the mixing of different starches (Fernando et al., 2001; Daramola & Osanyinlusi, 2006; Park et al., 2009).

Obanni and BeMiller (1997) and BeMiller (1997) attested to the fact that each starch is unique with unique functional properties, but much of that used industrially is modified before use, giving a wider range of useful products. Blending of starches has not been a common practice. Stute and Kern (1994) have patented blends of starches for use in pudding preparation. The patent claims that use of blends of unmodified pea and corn starches in ratios of 9:1 to 1:9 as gelling and texturing agents in the formulation of food products reduces syneresis. Liu and Lelièvre (1992) studied the melting transitions of blends of native wheat and rice starches. They found that, at starch concentrations <30 %, the differential scanning calorimetry (DSC) curves of the blends was the sum of the outputs for each individual component of the mixture. However, when starch concentrations were high, a nonadditive behavior was observed. Under these conditions, competition for water occurred and the starch with the lower gelatinization temperature (wheat) melted first (Liu & Lelièvre 1992). Jane and Chen (1992) blended amyloses and amylopectins from various botanical sources and reported synergistic effects on paste viscosities. When blends of a native or modified starch and a water-soluble gum are heated together in water, the general effects commonly seen were an apparent lowering of the pasting temperature as determined in a Brabender ViscoGraph or a Rapid Visco-Analyzer, a lower temperature of peak viscosity, a higher peak viscosity, a decreased rate of setback, a decreased gel strength, and a more rapid increase in the storage modulus of the paste. Obanni and BeMiller (1997) reported that mixing starches reduce

retrogradation, and also those with lower pasting temperature affect the properties of starch with higher gelatinization temperature.

Daramola and Osanyinlusi (2006) investigated the effect of active components of ginger roots on cassava starch. They found out that pasting properties of the ginger modified cassava starch showed high peak viscosities, low set back viscosities and low gelatinization time as compared to that of the native starch. The pasting profile of starch mixtures revealed that starches render mutual effects during pasting, more significantly when the amount of each component is similar (Eun et al., 2009).

Starch mixtures could have a place in the food industry where all natural products are desirable and in the food and other industries where reduction in the use of chemicals for modification is desirable. This is because new pasting properties can be generated by mixing starches of different botanical sources (Obanni & BeMiller, 1997; Daramola & Osanyinlusi, 2006; Zaidul et. al., 2007; Eun et al., 2009).

CHAPTER THREE

MATERIALS AND METHODS

This section describes the methodology. Three main experiments were carried out to achieve the stated objectives.

Morphological characterization

Collection of cassava planting materials

Forty-three distinct cassava landraces and cultivars were used. Thirty of them were obtained from the Plant Genetic Resources Research Institute (PGRRI), Bunso and the remaining thirteen from the University of Cape Coast (U.C.C.) Teaching and Research Farm. Three of the materials have been released as cultivars for the farmers.

Experimental Field and Field Layout

The field experiment was conducted on sandy loam soil at the Teaching and Research Farm of the School of Agriculture, U.C.C., Cape Coast. The soil has been described by Asamoah (1973) as Atabadze (equivalent to Ultisol in the United States Department of Agriculture, (USDA)) classification belongs to the Edina-Benya-Udu compound association, developed over Sekondian deposits. Cape Coast has a typical climate of the coastal savannah lowland characterized by an annual rainfall range of 800 to 1000 mm and mean monthly temperature of about 26.5 °C. The mean rainfall and the minimum temperature for the study area are shown on Table. 4.

A 430 m² land (43 m x 10 m) was ploughed, harrowed and divided into rows of 10.0 m in length with 1.0 m between rows in 2007 and 2008 major planting season. A total of 43 accessions were planted. Ten 20 cm-long cuttings were planted in single rows at a spacing of 100 cm within rows and 100 cm between rows. Ten plants for each accession were planted. An estimated total of 430 plants were cultivated.

Cultural Practices

The ploughed and harrowed field was lined and pegged before planting. Replacement of cuttings, which did not sprout, was not done as expected due to limited planting materials. The experiment was set out under rain fed conditions and weeding was done manually using a hoe or cutlass when necessary.

Data Collection

Both qualitative and quantitative characters from the germplasm collection were recorded. Incidence and severity of cassava mosaic viral disease was also evaluated. Most of the scoring was done in the field when the plants were two months old till harvesting at 12 months after planting (MAP). Morphological characterization was done based on qualitative and quantitative traits.

Table 4: Rainfall and Temperature Data for Cape Coast

Rainfall (mm) for Cape Coast												
YEAR	JAN	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	OCT	NOV	DEC
2007	0.0	0.7	68.8	44.1	71.6	146.1	222.8	44.7	117.4	133.1	21.3	10.3
2008	2.6	0.0	45.6	121.5	258.4	151.3	70.6	22.9	51.8	36.6	86.9	77.9
Minimum Temperature(⁰C) for Cape Coast												
YEAR	JAN	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	OCT	NOV	DEC
2007	19.8	21.1	21.4	20.9	21.1	20.1	20.3	20.8	20.7	21.1	21.4	21.3
2008	19.1	21.0	21.1	21.1	21.2	20.1	21.1	20.9	21.0	21.3	21.5	21.4

Source: Ghana Meteorological Agency

Observations and measurements were made on five out of ten plants for each accession. These plants were chosen after discarding two plants at the ends of the row. The standard cassava descriptors described by the International Board of Plant Genetic Resources (IBPGR) and adapted by the Genetic Resource Unit of International Institute of Tropical Agriculture (IITA, 1990) was used to describe the accessions for morphological traits. To score the colour traits, a colour chart (Munsell, 1977) was placed besides the trait concerned and the colour that marched the trait was used to describe it.

Leaf Characteristics

Leaf characteristics were taken at 6 MAP with the exception of the colour of first fully expanded leaf, which was taken at 3 MAP.

Colour of first fully expanded leaf (FLC)

The colour of first fully expanded leaf was recorded at three MAP and scored as follows: light green (3), dark green (5), green purple (7) and purple (9) as shown in Plate 1.

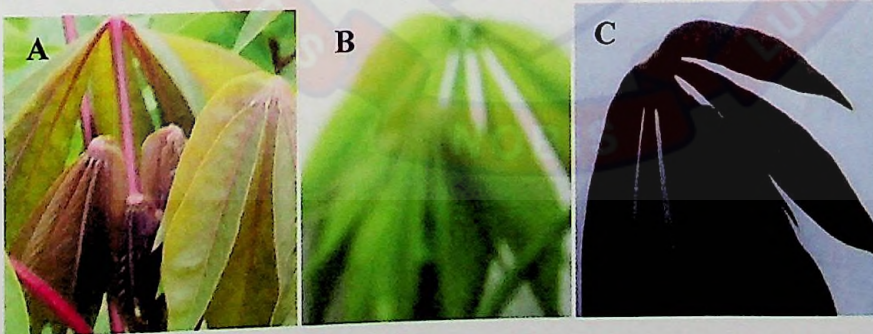


Plate 1: Various colour states of immature leaves of cassava; (A) Green purple, (B) Light green and (C) Purple

Leaf vein colour

The various colour states of the leaf vein recorded at 3 MAP are presented in Plate 2. The colour of the leaf vein was scored as follows: light green (3), Dark green (5), Green purple (7), Purple (9).

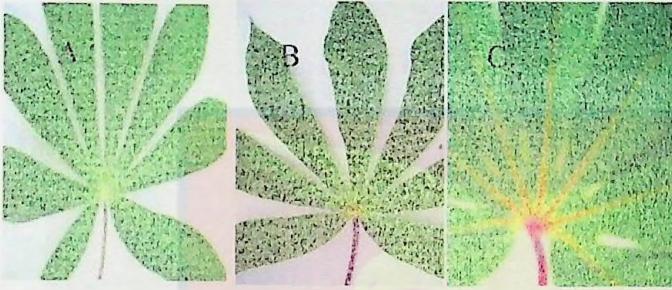


Plate 2: The colour states of leaf vein colour of the cassava germplasm, A) light green, B) green purple and C) purple

(c) Petiole colour

The colour of the petiole was recorded at 3 MAP and scored as follows: light green (3), Dark green (5), Green purple (7), Purple (9). The various colour states of the leaf vein are represented on plate 3.

(d) Central leaf lobe length (CLLL)

The central leaf lobe length was measured in five leaves using a metallic tape. The measurement was made along the mid-vein from the point where the lobes radiate from the petiole to the tip of the lobe. The average of the five leaves of each accession was taken as the central leaf lobe length.

(e) Width of the central leaf lobe (CLW)

The central leaf lobe width was also measured in five leaves. The widest portion was measured using a tape measure and the average of the five leaves of each accession taken as the central leaf lobe width. This is illustrated in Plate 4 below.



Dark green

Greenish purple

Purple

Plate 3: Variations in petiole colour of cassava



Plate 4: Procedure for measuring the central leaf lobe width of cassava germplasm

(f) Petiole length (PtL)

Petiole length was measured on fresh leaves. Five mature leaves were randomly selected from five plants of each accession. The petiole length of each leaf was measured from the point of attachment to the stem to the leaf base. Petioles with length between 5 and 14 cm were termed short, those with average length between 15 and 24 cm intermediate, while petiole lengths between 25 and 30 cm termed long.

(g) Young leaf pubescence

With the aid of a hand lens, the relative presence of hair on young leaves were observed and scored as follows: no pubescence (0); low pubescence (3); moderate pubescence (5) and high pubescence (7).

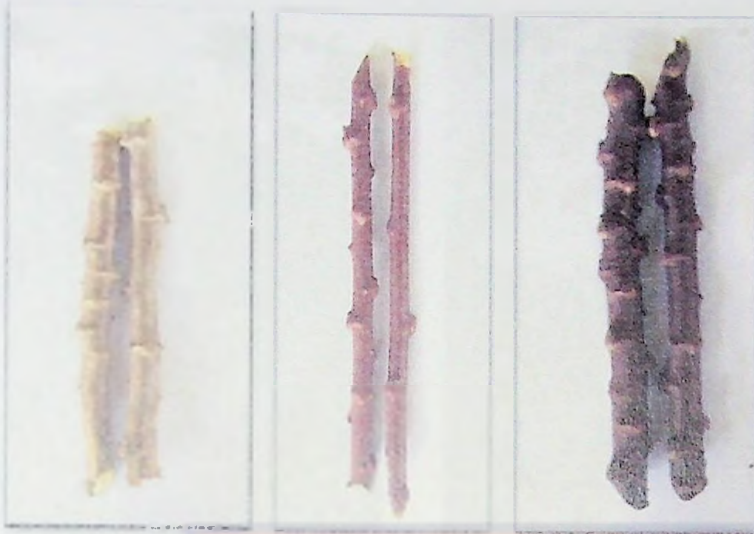
Stem Characteristics

(a) Stem colour (SC)

Stem colour was scored at 10 MAP. Three different stem colours were defined and scored as follows: silver green (1), light brown (2), and dark brown (3). The various character states of the stem colour observed are shown in Plate 5.

(b) Number of levels of branching (BrL)

The number of levels of branching per plant was counted at 12 MAP and recorded. Where there were two or more stems per stand the average number of branches was taken. Plate 6 shows the various branching levels encountered.



Silver

Light brown

Dark brown

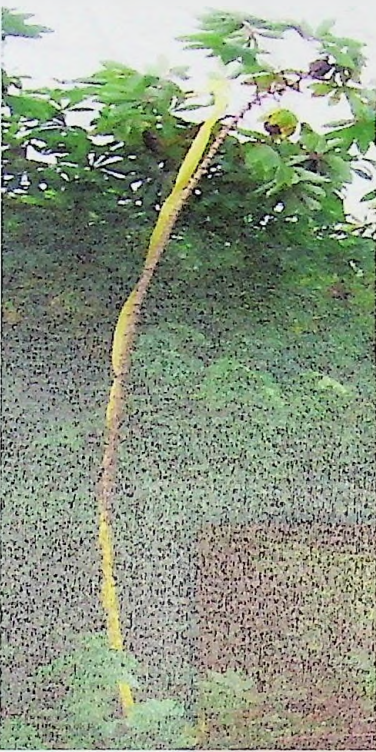
Plate 5: Colour states of stem of cassava accessions at 10 MAP

(c) Height of first apical branch (FBH)

Branching height was measured at 12 MAP from the soil level on the main stem to the point at which branching occurred, for two or more stems per stand the average of the branching heights were computed. Plate 7 illustrates how this trait was measured.

(d) Plant height (PH)

Plant height was measured at 12 MAP just before harvesting was done using five plants per accession. Plant height was measured from the soil level along the main stem to the highest point on the plant canopy.



No branching



One level of branching



Four levels of branching



Five levels of branching

Plate 6: Levels of branching among cassava accessions



Plate 7: Height of first apical branch

(e) Above ground fresh weight per plant (AGFW)

The above ground fresh weight is the total weight of the plant excluding the storage roots. This consists of the stem and the foliage. AGFW of all the sampled plants was determined after harvesting using a hanging scale. The total weight was divided by five to obtain the fresh shoot weight per plant.

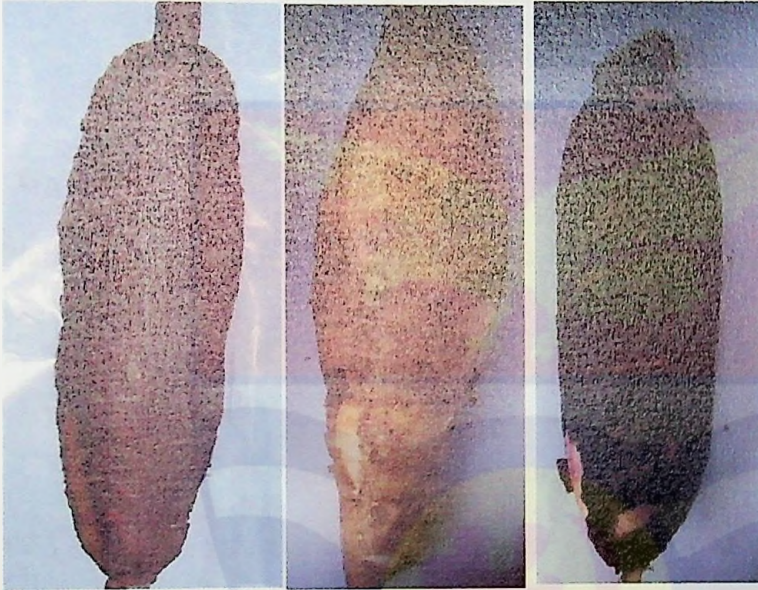
Tuber characteristics

(a) Number of tubers per plant

The total number of tubers per plant was determined by counting the number of tubers under each plant. Only wholesome tubers were counted. The average number of tubers for five plants was recorded to represent each accession.

(b) Outer surface colour of the root tuber

The colour of the outer surface of the root tuber was recorded immediately after harvesting. The tubers were first washed to remove all dirt and scored as follows: white or cream (1), light brown (2), and dark brown (3). The various colour states of the outer colour of the tuber are represented on plate 8.



Light brown

White/cream

Dark brown

Plate 8: Outer surface colour of the cassava root tuber

(c) Colour of outer cortex of the root tuber

The colour of the outer cortex was determined after carefully removing the outer sheath. The various colours were scored as follows: white or cream (1), yellow (2), pink (3), and purple (4). This is represented on Plate 9.



White

Yellow

Pink

Plate 9: Colour variation in outer cortex of cassava root tuber

d) Tuber fresh colour

The tuber fresh colour was determined after removing the peel/rind. Two different root tuber fresh colours were defined and scored as follows: white (1), and yellow (2). The various colour states observed are shown in plate 10.

(e) Root tuber length

Root tuber length was measured from the base of the tuber to the tip along the tuber. The measurement was made on five wholesome tubers and the average taken to represent the tuber length of the accession.

(f) Tuber diameter

The tuber diameter was determined by cutting the tubers at the middle. The diameter was measured from one end of the cut surface to the

other through the centre. Five tubers were randomly selected from five tubers and the average was taken to represent the tuber diameter for the accession. The variants were scored as follow: diameter with average length less than 5 cm were term as narrow, length between 5 and 10 cm were term intermediate, and length greater than 10 cm were termed as wide. They were scored as 1, 5 and 7 for narrow, intermediate and wide respectively



Yellow



White

Plate 10: Root tuber flesh colour encountered in the germplasm

(g) Tuber weight per plant

Tuber weight per plant was done by weighing the tubers. Tubers from five plants were bulked and placed in a sack. The sack was placed on

a hanging scale and the weight recorded. The average weight from the five plants, represent the tuber weight of the accession.

(h) Individual tuber weight (g)

The fresh root weight per plant obtained as described above was divided by the mean number of tubers per plant to obtain the individual tuber weight.

(i) Fresh root yield (t ha⁻¹)

The inter-row and intra-row spacing adopted were the same, that is, 1m apart and therefore the average plant population was 10,000 per hectare. Multiplying the number of plant stands by the mean fresh root weight (kg) and dividing by 1000 kg gave the fresh root yield in tonnes per hectare.

(j) Harvest index

Harvest index is the ratio of biological yield and economic yield of a crop. Biological yield represent the total dry matter accumulation of a plant system. Economic yield is the portion of the biological yield that constitute economic or agriculture value. The economic yield of cassava is the tubers. The harvest index for each plant was calculated as:

$$\text{Harvest Index} = \frac{\text{Weight of tuber}}{\text{Total plant weight}}$$

Molecular characterisation

Plant material

Forty three planting materials were use for the work. Thirty one (31) of them were obtained from Plant Genetic Resource Centre at Bunso

in the Eastern Region, while the remaining materials were obtained from the University of Cape Coast Cassava stock. Ten healthy cuttings were obtained for each accession and planted in a single row to sprout.

DNA extraction and purification

The youngest leaves were harvested when they were 14 days old for DNA extraction. DNA extraction was carried out at Crop Research Institute (CRI) Molecular Biology Laboratory, Fumensua. DNA was extracted by using a modified protocol by Dellaporta et al. (1983) and adopted by Crop Research Institute (CRI). This protocol consisted of cell lysis, DNA extraction and DNA precipitation and purification.

Cell lysis

Two hundred (200 mg) leaf samples were shock-frozen and ground into powder in liquid nitrogen using a Teflon pestle and 2-mL Eppendorf tube. To each tube, 800 μ L of lysis buffer was added and shaken several times until a homogeneous mixture was obtained. The mixture was incubated at 65 °C in a shaking water bath for 10 minutes and the tubes shaken by hand two times at 5-minute intervals to ensure uniform temperature within the tube.

DNA extraction

A solution of 5 M potassium acetate (400 μ L) was added and mixed gently by inversion of the tube. The incubated samples were immediately placed in ice for 30 minutes. The samples were centrifuged at 13000 rpm

for 10 minutes using microcentrifuge and the upper DNA-containing phase transferred into fresh tubes without disturbing the tube.

DNA precipitation and purification

An equal volume of cold isopropanol, which had been stored at -20°C , and $1/10^{\text{th}}$ of 3 M sodium acetate were added to each tube with the DNA-containing supernatant and the tube gently inverted a few times to precipitate the DNA. The samples were incubated at -20°C for 1 hour. The samples were centrifuged again at 13,000g for 10 minutes, the supernatant was discarded and the DNA pellet washed with cold 800 μL of 80% ethanol. The samples were centrifuged at 14,000 rpm for 5 minutes. This procedure was repeated until all colours were removed from the DNA extract.

The DNA pellets were air-dried on tissue paper spread on the laboratory bench for 30 minutes to remove the remaining ethanol droplets from the tube and redissolved in 500 μL of TE (Tris and EDTA) buffer. Four microlitres of RNase was added to the redissolved DNA samples and incubated at 37°C for 30 minutes to remove any RNA remaining. The DNA was purified with 250 μL of 7.5 M ammonium acetate, incubated on ice for 3 minutes and then centrifuge at 13,000 rpm for 5 minutes. The supernatant was transferred into a clean 1.5 mL eppendorf tube and 700 μL of cool isopropanol added, mixed by inversion and centrifuged at 13,000 rpm for 10 minutes. The DNA pellets were washed with 700 μL of 80 % ethanol, air-dried on tissue paper at room temperature re-dissolved in 100 μL of 1x TE buffer and stored at -20°C until required.

Determination of quantity and quality of DNA

The stored DNA samples were thawed, mixed thoroughly on a GENIE Vortex-2 (Scientific Industries, UK) and 5 μL added to 95 μL of sterile distilled water (SDW) in a 1 mL microcentrifuge tube and mixed well before reading the absorbance at 230, 260 and 280 nm using a Philips PU8720UV/VIS scanning spectrophotometer (Japan).

DNA quantity was calculated according to Weising et al. (2005) as:

$$\text{DNA } (\mu\text{g}/\mu\text{L}) = A_{260} \times 50,$$

where A_{260} is the absorbance at 260 nm

Thus the concentration of DNA in $\mu\text{g}/\text{mL}$ was calculated as:

$\text{DNA } (\mu\text{g}/\text{mL}) = [A_{260} \times 50] \times \text{DF}$ where DF is the dilution factor.

From the quantities of DNA calculated, the appropriate volumes were pipette into sample tubes and topped up with SDW to make concentration of 10ng/ μL used for polymerase chain reaction (PCR) amplifications. The ratio of the absorbance at 260nm to that at 280nm was used as determinants of DNA purity. Samples with ratios of 1.8 or greater were used for PCR amplification.

DNA samples were also run on 1 % agarose gels stained with ethidium bromide to assess their integrity. Each well contained a mixture of loading buffer (5 μL) and sample DNA (10 μL). The gels were run with 1x TAE buffer from the cathode to the anode with a constant voltage of 70 mV for 1.5 hours. They were visualized after electrophoresis with a UV transilluminator (UVP Inc., USA) and photographed with a cannon digital

(Canon, Power Shoot A620) camera. DNA samples with no visible shearing were then selected for subsequent PCR amplification.

PCR amplification of SSRs

Thirty-six highly polymorphic SSR markers (Table. 5), procured from Integrated DNA Technologies (Crolville, Iowa, USA), which are widely distributed in the cassava genome (Chavarriaga-Aguirre et al., 1998; Mba et al., 2000), were used in genotyping the accessions. PCR kits were obtained from Fermentas (IndustrialCol, South Africa). PCR reactions were conducted in a TECHNE TC-512 Thermocycler in a 10 μ L reaction mixture in 96-well plates. The mixture contained 1 μ L 10x PCR Buffer (Fermentas, Germany), 1 μ L 5mM dNTPs, 0.75 μ L each of the forward and reverse primers, 0.1 μ L of Super-Therm Taq polymerase (JMR-801, Fermentas, Germany), 3.4 μ L sterile distilled water (SDW) and 20 ng genomic DNA. The PCR programme consisted of an initial denaturation for 2 minutes (min) at 94 °C and then 30 cycles of denaturation for 30 seconds (s) at 94 °C, annealing at the appropriate temperature for each pair of primers for 30s, and extension at 72 °C and then put on hold at 4 °C at infinity (∞). The amplified products were stored at -20 °C until they were needed to run gels.

Table 5: Cassava SSR markers used for studying genetic diversity

Locus	Type of Repeat	Primer	Temp (°C)
SSRY4-R	(GA) ₁₆	ATAGAGCAGAAGTGCAGGCG	57.0
SSRY4-F		CTAACGCACACGACTACGGA	
SSRY9-R	(GT) ₁₅	ACAATTCATCATGAGTCATCAACT	53.6
SSRY9-F		CTAACGCACACGACTACGGA	
SSRY19-R	(CT) ₈ (CA) ₁₈	TGTAAGGCATTCCAAGAATTATCA	53.3
SSRY19-F		TCTCCTGTGAAAAGTGCATGA	
SSRY20-R	(GT) ₁₄	CATTGGACTTCCTACAAATATGAAT	52.7
SSRY20-F		TGATGGAAAGTGGTTATGTCCTT	
SSRY21-R	GA(26)	TGATGGAAAGTGGTTATGTCCTT	52.9
SSRY21-F		CCTGCCACAATATTGAAATGG	
SSRY51-R	(CT) ₁₁ CG(CT) ₁₁ (CA) ₁₈	AGGTTGGATGCTTGAAGGAA	55.9
SSRY51-F		GGATGCAGGAGTGCTCAACT	
SSRY59-R	(CA) ₂₀	GCAATGCAGTGAACCATCTTT	53.9
SSRY59-F		CGTTTGTCTTTCTGATGTTC	
SSRY64-R	(CT) ₁₃ CG(CT) ₆	ACAAGTCGTATATGTAGTATTCA	55.1
SSRY64-F		GCAGAGGTGGCTAACGAGAC	
SSRY69-R	(CT) ₁₈ ATT(AT) ₂ (N) ₇	CGATCTCAGTCGATACCCAAG	55.0
SSRY69-F		ACTCCGTTGCAGGCTTA	
SSRY82-R	(GA) ₂₄	GTGACAATTTTCAGATAGCTTCA	52.8
SSRY82-F		ACCATCGGCATTAAACTTTG	
SSRY100-R	(CT) ₁₇ TT(CT) ₇	TCCTTGCCTGACATTTTGC	53.7
SSRY100-F		TCGCAGAGTCCAATTGTTG	
SSRY101-R	(GCT) ₁₃	GAGAATACCACCGACAGGA	54.9
SSRY101-F		CAGCAGCAATCACCATTTT	
SSRY103-R	(GA) ₂₂	GAGAAGGAAACTGCTTGAC	55.6
SSRY103-F		AGCAAGACCATCACCAGTTT	
SSRY47-R	(CA) ₁₇	GAGCACCTTTGCTGAGTT	54.6

Table 5 continued

SSRY47-F		TGGAACAAAGCAGCATCAC	
SSRY151-R		GTGGAAATAAGCCATGTGATG	53.1
SSRY151-F		CCATAATTGATGCCAGGTT	
SSRY155-R		GTTGATAAAGTGGAAAGAGCA	57.6
SSRY155-F		CTCCACTCCCGATGCTCGC	
SSRY175-R	GA(38)	GACTAGCAGACACGGTTTCA	54.7
SSRY175-F		CTAACAGTCCAATAACGATAAG G	
SSRY179-R	(GA) ₂₈	AGGCTCAGGTGAAGTAAAGG	55.2
SSRY179-F		CGAAAGTAAGTCTACAATTTTCT AA	
SSRY181-R	(GA) ₂₂ (G) ₃ C(GA) ₃ GGAA(GA) ₄	GTAGATCTGGATCGAGGAGG	54.8
SSRY181-F		AATCGAAACCGACGATACA	
NS911-F		GTTGTTCAGACGATGTCCAA	53.2
NS911-R		TGAAGCAGTTATGAACCGT	

Running of gel

Horizontal polyacrylamide gel electrophoresis (hPAGE) was used for running the samples. 6 X Orange DNA loading dye [10 mM Tris-HCl (pH 7.6), 0.15 % orange G, 0.03 % xylene cyanol FF, 60 % glycerol, 60 mM EDTA] was used. The loading dye contained two dyes, orange G and xylene cyanol FF for visual tracking of DNA migration during electrophoresis. The presence of glycerol in the solution ensures that the same sample forms a layer at the bottom of the well; the EDTA included in the solution binds divalent metal ions and inhibits metal dependent nucleases. One volume of the dye was added to 5 volumes of DNA

sample. After initial denaturing at 95 °C for 5 min using the PCR machine, 6 µL of sample (or DNA ladder in the first well) was loaded in each well of a 41-well 5 % polyacrylamide gel. A 50 bp DNA molecular weight marker (Fermentas, Germany) was run in each gel as a reference to estimate the size of specific DNA bands in the PCR-amplified products. Gels were run at 300V for 2 hours using a Baid and Tatlock Nucleic Acid Electrophoresis Cell and power pack and 1x TBE as running buffer. The products were visualized by ethidium bromide staining method. The gel (attached to the back glass plate using bind-Silane) was stained in 500 mL TE buffer containing 25 µL of ethidium bromide for 50 min. The stained gel was washed in 500mL of TE buffer to get rid of excess ethidium bromide on the gel, viewed using a UV transilluminator (Uvitech, UK) and photographed with a digital camera (Canon, Power Shoot A620).

Screening for mosaic resistance

Field screening/morphological screening

The 43 cassava accessions were evaluated in two growing seasons, 2007/2008 and 2008/2009 on the Teaching and Research Farm, University of Cape Coast. The site is in the coastal savannah zone with a ferric luvisol soil type and is a high pressure site for the disease. Population of white flies, disease incidence and severity were assessed.

Whitefly population

Whiteflies (*Bemisia tabaci*) are the vectors of African cassava mosaic virus (ACMV) disease on cassava plants. Determining their

population on cassava plants would therefore aid in assessing the relationship between numbers of whiteflies and the severity and incidence of ACMV disease infection.

Direct counts of adult whiteflies on the crop were made (Mound, 1965; Hill, 1968; Fargette et al., 1985; Abdullahi et al., 2003). Whitefly counting was usually done between 0600 and 0800 hours when the environment was cooler and whiteflies were relatively immobile than later in the day (Fauquet et al., 1987). Adult whitefly populations on the five topmost expanded leaves of the selected cassava cultivars were counted (Otim-Nape et al., 2005; Ariyo et al., 2005). No matter how luxuriant a genotype was in growth and canopy spread, whitefly count was often carried out on the five topmost expanded leaves. Five plant stands that were affected by ACMV disease were randomly selected for each cassava genotype. On each plant, leaves were carefully turned over and the number of adult whiteflies under the leaves surface were counted and recorded. The mean number of whiteflies per 5 top leaves was then computed. The counts were done one month after planting and were repeated at the third and sixth months after planting.

African Cassava Mosaic Virus (ACMV) disease score

Plants infected by ACMV disease have their leaves reduced in size, misshapen and twisted, with chlorotic areas separated by green areas. Leaflets may show a nearly uniform mosaic pattern while others may show mixed mosaic patterns.

Scoring for ACMV disease was done one, three and six months after planting. The following ordinal scoring system (IITA, 1990; Ariyo et al., 2005) was used.

1. no symptoms observed.
2. mild chlorotic pattern on entire leaflets or mild distortion at base of leaflets appearing green and healthy.
3. strong mosaic pattern on entire leaf, and narrowing and distortion of lower one-thirds of leaflets.
4. severe mosaic distortion of two-thirds of leaflets and general reduction of leaf size.
5. severe mosaic distortion of four-fifths or more of leaflets, twisted and misshapen leaves.

Five plants for each accession were scored and the mean ordinal score computed.

Plants with a mean CMD scores of “1” were then classified as highly resistant (HR), those with a score of “2” were resistant (R), those with a score of “3” were classified as susceptible (S) and those with scores of “4” and “5” were classified as highly susceptible (HS).

Laboratory screening for various strains of cassava mosaic viruses

Collection of cassava leaf samples

Young cassava leaves from 43 accessions were collected from both mosaic infected plants and uninfected plants at the School of Agriculture Teaching and Research Farm, UCC. The leaves were packed into a zipped polythene bag and transported to Molecular Biology and Biotechnology

laboratory UCC. The total DNA was isolated using the same procedure described above. The DNA was stored at -20°C .

PCR amplification of Cassava Mosaic primers

The 43 samples were tested for presences or absences of cassava mosaic disease using primers that could detect African Cassava Mosaic Virus (ACMV), East African Cassava Mosaic Virus (EACMV) and East African Cassava Mosaic Virus Ugandan variant (EACMU-Ug). In all six pairs of primer sequence designed earlier by Harrison et al., 1997 and Zhou et al., 1997, were obtained (Table 6). The PCR reactions were conducted using Applied Biosystems Thermal Cycler version 2.08 (2720 Thermal Cycler) in 96 well plates. The reaction mixture composed of 10 μL which consist of AccuPower PCR Premix, genomic DNA, sterile distilled water (SDW) and primers. The PCR mixture contained 9 μL of PCR premix and primers and 1 μL 10 ng μL^{-1} genomic DNA.

Table 6: Nucleotide sequences of DNA primers used in polymerase chain reaction for the detection of cassava mosaic begomovirus

Virus	Name of primer	Sequence (5' to 3')
ACMV1	ACMV-F1	TTC AGT TAT CAG GGC TCG TAA
	ACMV-R1	GAG TG AAG TTG ACT CAT GA
ACMV2	ACMV-F2	GTG AGA AAG ACA TTC TTG GC
	ACMV-R2	CCT GCA ATT ATA TAG TGG CC
ACMV-AL	ACMV-AL1/F	GCG GAA TCC CTA ACA TAA TC
	ACMV-ARO/R	GCT CGT ATG TAT CCT CTA AGG CCT
ACMV3	ACMV-1	GCTC AAC TGG AGA CAC ACT TG
	ACMV-2	CCT GCA ACA TAC TTA CGC TT
EACMV/EACMV	UV-AL3/F	TAC ACA TGC CTC RAA TCC TG
	UV-AL1/F1	CTC CGC CAC AAA CTT ACG TT
EACMV-Ug	UV-AL1/F1	TGT CTT CTG GGA CTT GTG TG
	ACMV-CP/R3	TGC CTC CTG ATG ATT ATA TGTC

The PCR programme consisted of an initial denaturation for 4 minutes at 94 °C and then 35 cycles of denaturation for 30 seconds (s), at 94 °C, annealing at 50 °C (it could be 50 °C to 60 °C) for 30 s, depending on the annealing temperature of the primer and extension at 72 °C and for 90 °C and then put on hold at 4 °C at infinity (∞). The PCR products were separated by electrophoresis in a 1 % agarose gel at 100 volts for about 1.5 h. The gel was stained with ethidium bromide and viewed under UV light. The amplified products were kept at -20 °C until they were needed to run gels.

Screening for source of resistance

Plant samples that did not show presence of any of the strains of cassava mosaic virus were further amplified with specific SSR primers that are associated with CMD2 gene, which confers resistance to CMD. The four primer pairs were SSRY28, NS156, NS169 and RME1. DNA amplification, running of the gel and staining were the same as those described above.

Starch Extraction

Five hundred gram of root tubers was used. The pulp cut into small cubes. Each sample was milled with excess distilled water in a Kenwood blender for about 5-7 minutes to produce a fine milky pulp. The starch slurry was then filtered through a cheese cloth, folded in duplicate, into a plastic container. More water was added and squeezed until no more milky starch came out. The distillate was allowed to stand for about four hours to settle after which the supernatant was drained away. The pure white starch

in the plastic container was dried in the sun for about 3-4 days to a constant weight after which it was allowed to cool and then weighed using analytical weighing scale. The starch samples were kept in zipped polythene bags.

Calculation of Starch content (%)

The percentage starch was estimated as:

$$\% \text{ Starch} = \left(\frac{\text{Starch weight}}{\text{Fresh weight of cassava}} \right) \times 100$$

Starch yield (t ha⁻¹)

Starch yield (t ha⁻¹) of each accession was computed by multiplying the fresh root weight (t ha⁻¹) of a given accession by its percentage starch content.

Functional properties of starch

The functional properties of starch, that is, the swelling volume, swelling power and solubility of starch, were determined based on a modification of the method of Leach et al. (1959). One gram of sample was transferred into a weighed graduated 50 mL centrifuge tube. Distilled water was added to give a total volume of 40 mL. The suspension was stirred just sufficiently and uniformly with a stirrer avoiding excessive speed, in order not to cause fragmentation of starch granules. The suspension was heated to 85 °C in a water bath (Grant instruments, England LTD), shaking gently to ensure that the starch granules remain in suspension until gelatinization occurs (5 minutes). The gelatinized sample was held at 85 °C in the water bath for 30 minutes. The sample was cooled

to room temperature under running water and then centrifuged for 15 minutes at 2200 rpm. It was then centrifuged for 15 min at 2200 rpm in a centrifuge (Gallenkamp, England). The solubility was determined by evaporating the supernatant in a hot air oven (BS Gallenkamp, England) and weighing the residue. The swelling volume was obtained by directly reading the volume of the swollen sediment in the tube. The sedimented paste was weighed. Determinations were done in triplicate. The % solubility and swelling power were then calculated as follows;

(a) Swelling volume

Swelling volume was obtained directly by reading the volume of the swollen sediment in the tube.

(b) Solubility

The soluble starch was decanted carefully into a cleaned and weighed glass crucible and evaporated in an oven at 90 °C till constant weight was achieved. The percentage solubility was then calculated from the dried residue,

$$\% \text{ Solubility} = \left(\frac{\text{Weight of soluble sample}}{\text{Weight of sample (dry basis)}} \right) \times 100$$

(c) Swelling power

Swelling power was determined by weighing the sediment and expressing swelling power as the weight (g) of swollen sediment over gram

dry starch, that is, swelling power was determined using the following relationship:

$$\text{Swelling Power} = \frac{W1}{W2 - W3}$$

Where:

W1= Weight of sedimented paste

W2 =Weight of initial sample (dry basis)

W3 = Weight of sample which dissolves away

Pasting characteristics

The pasting characteristics of the starch samples were determined using the Brabender Viscograph instrument. First, the moisture content of each sample was determined using an electronic moisture meter. The value of the moisture content of a sample was fed into the software of the Brabender Viscograph and the instrument automatically indicated the weight of starch sample to be used and the quantity of distilled water to be added to make starch slurry (suspension).

The slurry was then put into an aluminium canister of the instrument and heated at a rate of $1.5\text{ }^{\circ}\text{Cmin}^{-1}$ by means of a thermostat at a speed of 75 rpm. The start temperature was $50\text{ }^{\circ}\text{C}$. When the suspension reached $95\text{ }^{\circ}\text{C}$, it was held constant for 15 minutes (first holding period) while being continuously stirred. The paste was then cooled down to $50\text{ }^{\circ}\text{C}$ at a rate of $1.5\text{ }^{\circ}\text{C min}^{-1}$, and held at this temperature for another 15 minutes (second holding period).

At the end of the process which took 1 hour 30 minutes, the following records were read from the Viscograph printed out by the instrument:

- (a) Pasting temperature ($^{\circ}\text{C}$)
- (b) Pasting time (in minutes)
- (c) Peak viscosity (in Brabender Units [BU])
- (d) Peak temperature ($^{\circ}\text{C}$)
- (e) Peak time (minutes)
- (f) Viscosity at 95°C (BU)
- (g) Viscosity after 15 minutes at 95°C (BU)
- (h) Viscosity at 50°C (BU)
- (i) Viscosity after 15 minutes at 50°C (BU)
- (j) Paste stability at 95°C (BU)
- (k) Paste stability at 50°C (BU)
- (l) Setback viscosity (BU)
- (m) Breakdown viscosity (BU)

Paste stability at 95°C and paste stability at 50°C were computed as the difference between viscosity at 95°C and viscosity after 15 minutes at 95°C ; and the difference between viscosity at 50°C and viscosity after 15 minutes at 50°C respectively.

Ease of cooking was estimated according to Sanchez et al. (2009). It is estimated by subtracting pasting time from peak time.

Data Analysis

Morphological Data

Twenty descriptors were recorded from the cassava germplasm in the study area and used for descriptive statistics and correlation coefficients analysis performed using SPSS statistical software.

Coefficient of variation was calculated as follows:

$$CV = \frac{\sigma}{x} \times 100$$

σ is the standard deviation and

x is the general mean of the trait being investigated.

The relationship between each two set of trait was studied using correlation analysis.

Factor analysis was performed on all morphological traits. Factor analysis summarizes data into a few dimensions by condensing a large number of variables into a smaller set of latent variables or factors. Selected traits were subjected to principal component analysis to know amount of variance each traits contributes. Principal component analysis of the traits was employed to examine the percentage contribution of each trait to total genetic variation.

Molecular Data

Estimation of genetic diversity SSR markers, which resulted in complex patterns (i.e. more than 2 alleles) were excluded to maintain a strict di-allelic model of inheritance (Fregene et al., 2003). Twenty markers that gave distinct diallelic patterns were chosen for gene diversity analysis (Table 10). To determine the relationship among accessions based

on a hierarchical cluster analysis, the alleles were recorded as bands and scored as 1 or 0 for present and absent respectively. The data in this form were used to calculate genetic distances between pairs of cassava accessions from comparisons of the band scores. Then using the unweighted pair-group mean average (UPGMA) cluster method of Nei's genetic distances (Sneath & Sokal 1973), a dendrogram of genetic similarity was generated. The genetic distances and dendrogram were computed with the PowerMarker computer programme, version 3.25 (Liu & Muse 2005). To estimate genetic diversity among the accessions, SSR loci were recorded as diploids with single bands taken to indicate the presence of two identical alleles. Genetic diversity among accessions partitioned into the cluster groups generated from the genetic similarity analysis was estimated according to Liu and Muse (2005). Genetic diversity was estimated using five statistics averaged over loci; the percentage of polymorphic information count (PIC); the mean number of alleles per locus or allelic richness (A); the average observed heterozygosity (H_o); and the average gene diversity (H_e) were computed, according to Nei (1983).

CHAPTER FOUR

RESULTS

Variation in qualitative characters

Colour of first fully expanded leaf (FLC)

The definitive grades recorded for the colours of the first fully expanded leaf were light green, green purple, and purple (Figure 4). The first leaf colour was characterized predominantly by light green with relative frequency of about 41.9 % and 34.9 % for dark green which is the second dominating colour character state of the trait. The cultivars with green purple and purple had smaller percent of 11.6 % respectively.

Leaf vein colour

The leaf vein colours present were light green, green purple and purple. The distribution of leaf vein colour is presented in Figure 5. Accessions with purple leaf vein predominated at frequency of 41.9 %, followed by light green with a percentage of 34.9 %, whilst accessions with green purple leaf vein colour recorded the least frequency of 23.2 %.

Petiole colour

Figure 6 shows the distribution pattern of petiole colour among the total accessions. Three classes were observed light green, green purple and purple. With 69.7 % accessions with purple petiole colour predominated

among the total accessions. Cultivars with light green petiole colour had the lowest frequency of about 9.4 %.

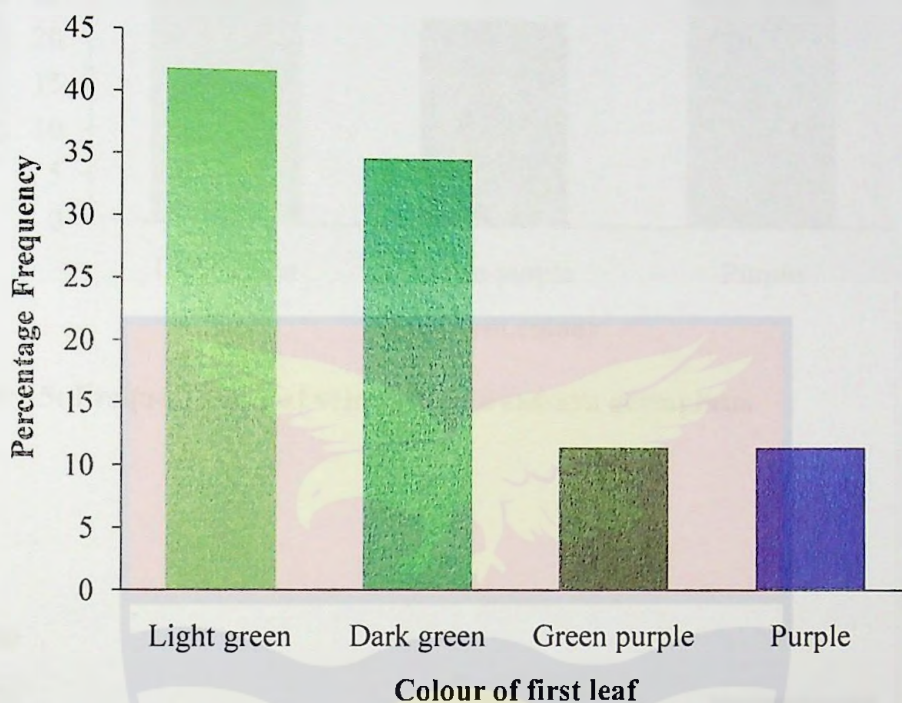


Figure 4: Frequency of first leaf colour in cassava germplasm

Petiole length was scored on a scale of short (5-14 cm), intermediate (15-25 cm) and long (24-30 cm). Distribution of petiole length for the accessions is presented in Figure 7. Frequency of accessions with long petiole was the highest among the total accessions with a percent frequency of 51.2 %. This was followed by medium petiole with a percentage frequency of 46.5 %. Accessions with short petiole length constitute only 2.3 %.

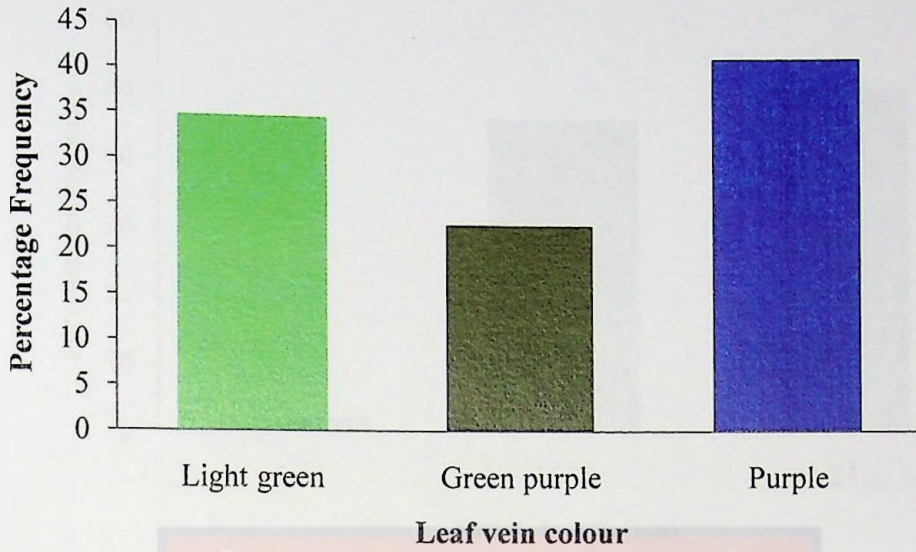


Figure 5: Frequency of leaf vein colour in cassava germplasm

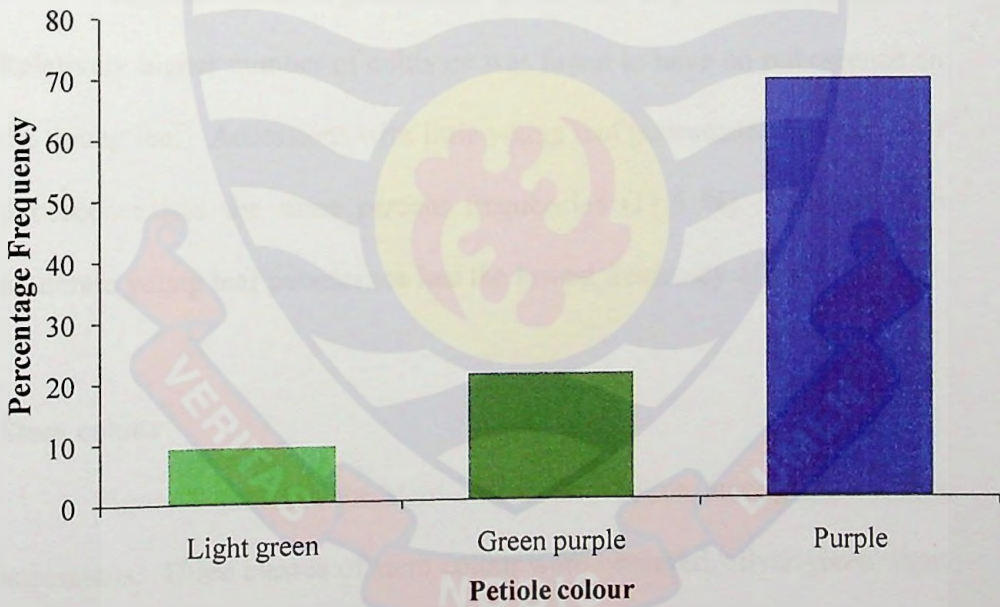


Figure 6: Frequency of petiole colour in cassava germplasm

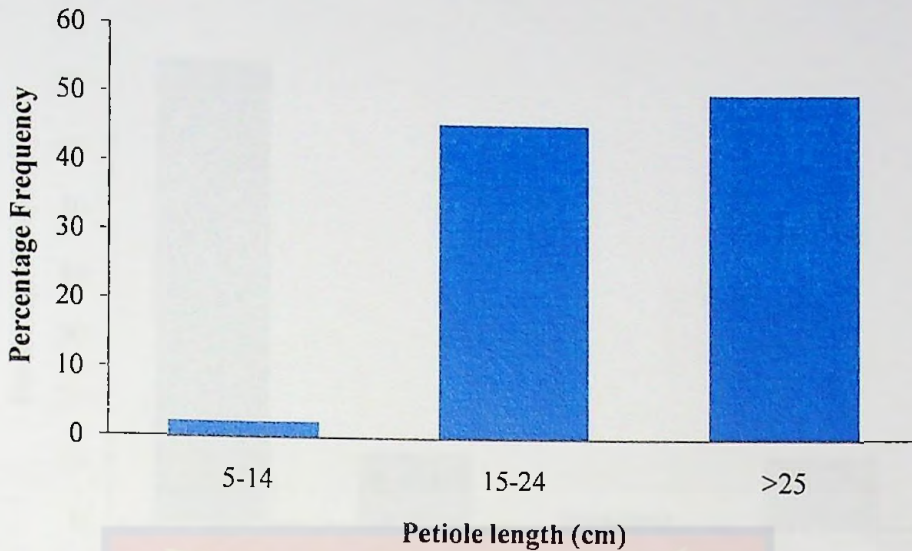


Figure 7: Frequency of petiole length in cassava germplasm

Pubescence of young leaf

The distribution of pubescence of first leaf is presented in Figure 8. Relatively higher number of cultivars was found to have no pubescence on the young leaf. Accessions with little young leaf pubescence and high leaf pubescence had the same percent frequencies (11.6 %). Cultivars with moderate young leaf pubescence had the lowest frequency of about 2.3 %.

Stem colour

Figure 9 presents the stem colour distribution among the cassava accessions. Three classes of stem colour were observed, silver green, light brown and dark brown. Among the total accessions light brown stem colour predominated with a percentage frequency of 43 %. The second predominant colour observed was dark brown with a percentage frequency of 35 %. Silver green stem colour was the least colour recorded (22 %).

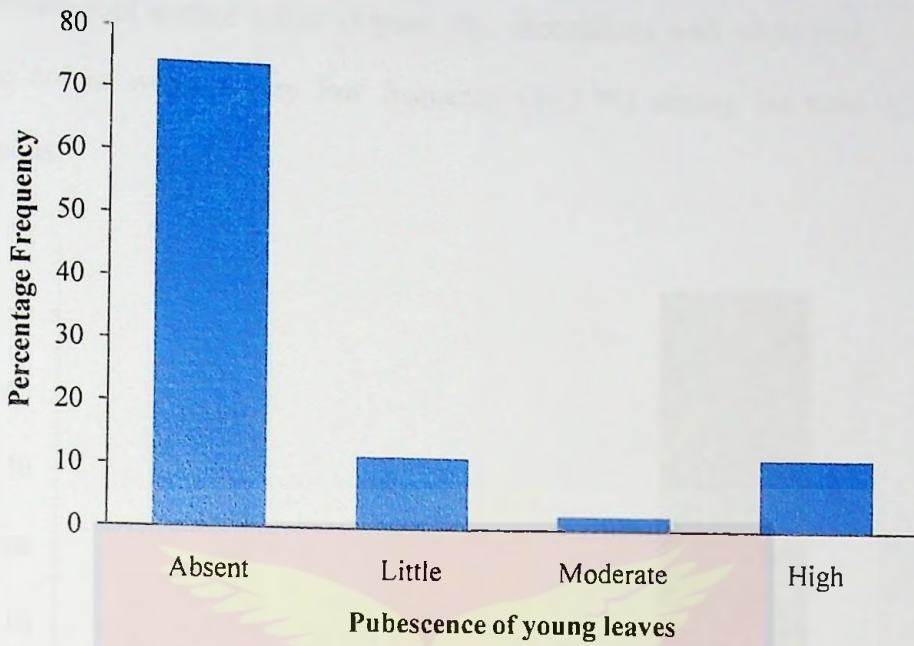


Figure 8: Frequency of pubescence of young leaf in cassava germplasm



Figure 9: Frequency of stem colour in cassava germplasm

Storage root surface colour

Three classes of root surface colour were observed. These are white/cream, light brown and dark brown. Over 50 % of the accessions had

dark brown root surface colour (Figure 10). Accessions with white root surface colour were at very low frequency (16.3 %) among the total accessions.

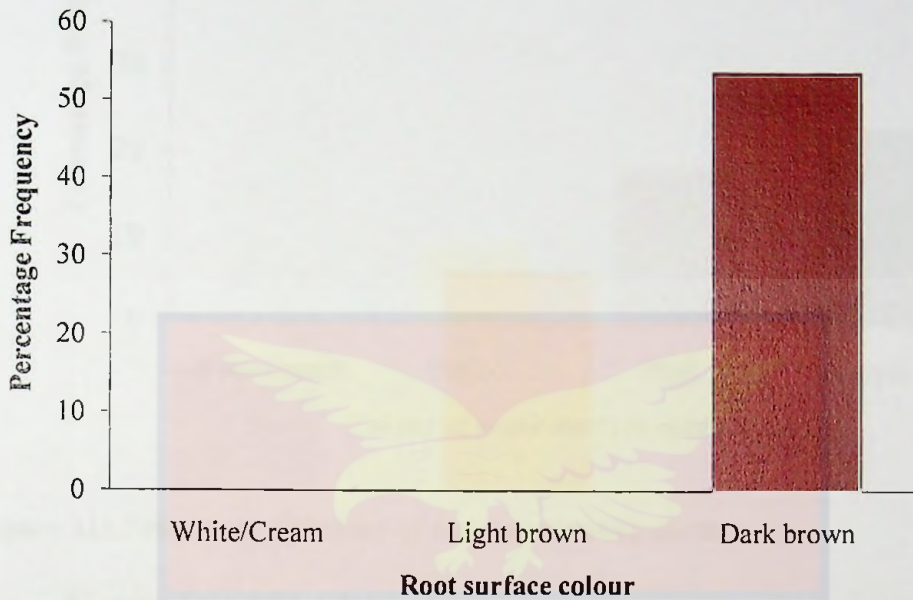


Figure 10: Frequency of root surface colour in cassava germplasm

Colour of outer root tuber cortex

Figure 11 presents the outer surface root cortex colour distribution among the cassava accessions. Four classes of colour were observed, white, pink, yellow and purple. Among the total accessions white/cream outer surface root cortex colour predominated with a percentage frequency of 48.8 %. The second predominant colour observed was purple with a percentage frequency of 23.3 %. Yellow outer surface root cortex colour was the least colour recorded (9.3 %).

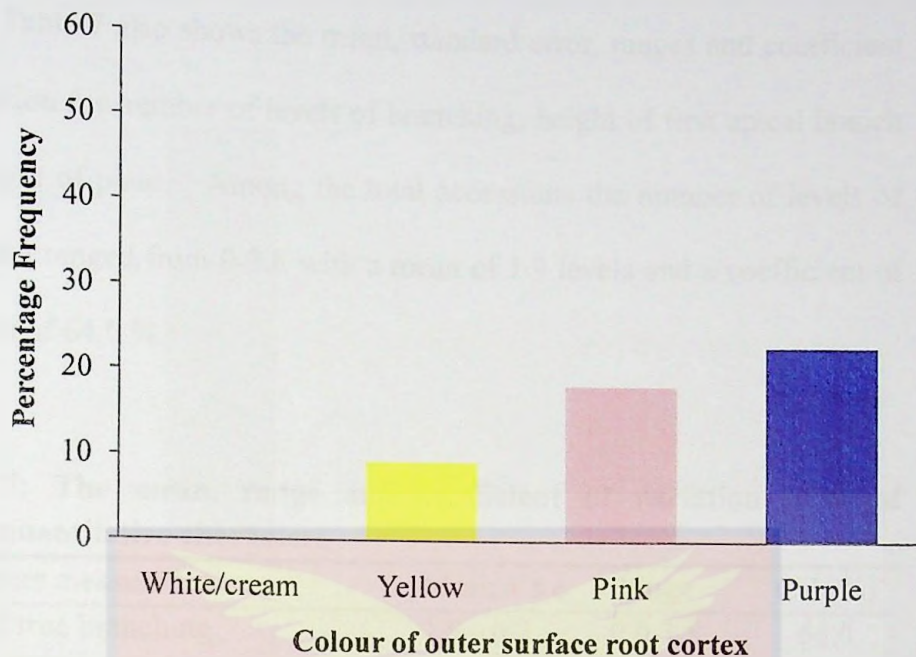


Figure 11: Frequency of colour of outer root tuber cortex

Storage root pulp colour

Two classes of root pulp colour were observed. Over 97.7 % of the accessions had white/cream root pulp colour. Accessions with yellow root pulp colour were at very low frequency (2.3 %) among the total accessions.

Variation in quantitative characters

The mean, standard error, ranges and coefficient of variation for the quantitative characters are shown in Table 7. Among the total accessions the petiole length ranged between 9.6-34 cm with a mean of 22.5 cm and a coefficient of variation of 4.7 %. Length of central leaf lobe ranged between 9.9-25.3 cm with a mean of 19.2 cm and a coefficient of variation of 23.2 %. Among the total accession, central leaf lobe width ranged between 2.9-9.3 cm with a mean of 5.7 cm and coefficient of variation of 23.2 %.

Table 7 also shows the mean, standard error, ranges and coefficient of variation for number of levels of branching, height of first apical branch and height of plant. Among the total accessions the number of levels of branching ranged from 0-3.8 with a mean of 1.9 levels and a coefficient of variation of 64.0 %.

Table 7: The mean, range and coefficient of variation (CV) of quantitative characters

Parameters measured	Mean \pm s.e	Range	CV (%)
Level of true branching	1.9 \pm 0.2	0.0-3.8	64.0
Height of first apical branching (cm)	89.9 \pm 7.6	0.0-192.0	55.3
Level of lateral branching	2.8 \pm 0.4	0.0-8.8	85.5
Height at first lateral branching (cm)	29.6 \pm 3.7	0.0-108.8	81.7
Central lobe length (cm)	19.2 \pm 0.7	9.9-25.3	23.2
Central lobe width (cm)	5.7 \pm 0.2	2.9-9.3	23.2
Petiole length (cm)	22.5 \pm 0.9	9.6-34.3	4.7
Plant height (cm)	297.1 \pm 8.9	115-397	19.6
Number of rotten storage root/plant	0.2 \pm 0.05	0.0-1.2	58.8
Storage root length (cm)	34.4 \pm 1.7	5.0-50	23.3
Storage root diameter (cm)	6.6 \pm 0.4	2.0-13.7	36.3

The height at which the first apical branch occurs ranged from 0.0-192 cm, for the total accessions with a mean of 89.9 cm and coefficient of variation of 55.3 %. The plant height for all the accessions studied ranged from 115.0-397.0 cm with a mean of 297.1 cm and a coefficient of variation of 19.6 %.

The level of lateral branching and height at which it occurred was considered. The range for the level of lateral branching was 0.0-8.8 with a mean of 2.8 and a coefficient of variation of 85.5 %. Among the total accessions the height at which lateral branch occurred ranged between 0.0-108.8 cm with a mean of 29.6 cm and a coefficient of variation of 81.7 % (Table 7).

Associations among quantitative agronomic traits

Associations among the character states were studied using correlation analysis. Associations among quantitative agronomic characters are presented in Table 8. There was no significant correlation between level of true branching and the rest of the quantitative traits studied

Plant height recorded a highly significant and positive correlation with top weight ($r = 0.80$; $p < 0.01$), root weight per plant ($r = 0.59$; $p < 0.01$) and root length ($r = 0.47$; $p < 0.01$). Central leaf lobe length correlated positively and significantly with central leaf lobe width ($r = 0.70$; $p < 0.01$), petiole length ($r = 0.76$; $p < 0.01$), top weight ($r = 0.43$; $p < 0.01$), root weight per plant ($r = 0.51$; $p < 0.01$) and harvest index ($r = 0.43$; $p < 0.01$).

Central leaf lobe width significantly and positively correlated with petiole length ($r = 0.75$; $p < 0.01$), top weight ($r = 0.67$; $p < 0.01$) and root weight per plant ($r = 0.62$; $p < 0.01$). It also correlated positively and significantly with the harvest index ($r = 0.33$; $p < 0.05$).

Top weight had significant and positive correlation with root weight per plant ($r = 0.72$; $p < 0.01$) and root length ($r = 0.52$; $p < 0.01$). Root weight on the other hand, correlated significantly and positively with starch yield ($r = 0.46$; $p < 0.01$), harvest index ($r = 0.73$; $p < 0.01$), root length ($r = 0.65$; $p < 0.01$) and root diameter ($r = 0.43$). Percentage starch yield had significant and positive correlation with harvest index ($r = 0.45$; $p < 0.01$) and root length ($r = 0.48$; $p < 0.01$). Harvest index correlated significantly and positively with root length ($r = 0.52$; $p < 0.01$) and root diameter ($r = 0.306$; $p < 0.05$).

Genetic diversity analysis at morphological level

Cluster Analysis

Cassava accessions exhibited greater degree of genetic variation for nineteen different morphological both quantitative and qualitative traits observed. All the morphological traits recorded were subjected to factor and principal component analysis. Principal component 1, 2 and 3 accounted for 46.6 %, 14.7 % and 11.4 % of total variance, respectively. Thus, the maximum percentage of variance that contributed to the total variability was 46.6 % (Table 9).

Similarity index for the recorded morphological traits among the cassava accessions was worked out by calculating the Nei's similarity coefficient. Nei's coefficient matrix was used to construct the hierarchical clusters and resulting dendrogram as presented in Figure 12. The similarity coefficient ranged from 0.0 to 0.40. The forty-three cassava accessions were grouped into two broad clusters, however, at similarity coefficient of 0.15 they were further divided into four clusters. Among these different clusters, the cluster size varied from 5 for cluster I to 18 for cluster IV. Clusters II and III had 10 members each, while the highest number of accessions was found in cluster IV. Adehye and Capevars cassava varieties were closely related in cluster (III)

Seven of the traits (stem colour, colour of young leaf, colour of root cortex, leaf vein colour, petiole length, yield and root surface colour) showed more variation among the varieties than other traits (pubescence of young leaves, plant height, petiole length, pulp colour and root diameter).

Genetic diversity analysis using SSR markers

Twenty out of 36 primers gave polymorphic bands; the remaining primers were either monomorphic or failed to amplify any product and therefore were not considered for further analysis. The size of amplified alleles ranged from 75 to 350 bp as shown in Table 10 and Plate 11. The SSR primers were able to distinguish all the 43 accessions used for the study. The four most polymorphic primers were SSRY175, SSRY101, SSRY20 and SSRY100. The number of alleles per SSR loci among the 43 cassava genotypes ranged from two to nine alleles. The mean number of alleles was 4.91 alleles.

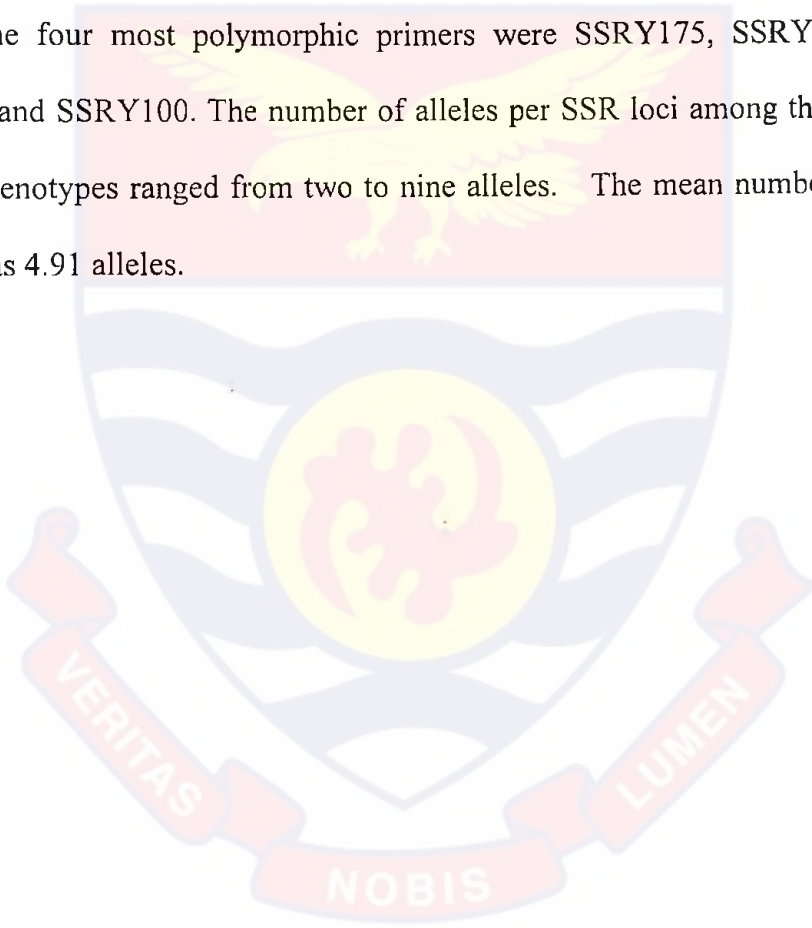


Table 8: Correlation coefficient for quantitative traits

	B	C	D	E	F	G	H	I	J	K	L	M	N	O
A	0.089	-.345(*)	-0.065	0.022	0.221	0.11	0.034	0.13	0.082	0.167	0.05	0.187	0.185	0.066
B		0.216	.315(*)	.339(*)	0.105	0.163	.317(*)	0.269	0.22	0.047	0.105	-0.048	0.098	-0.231
C			.372(*)	.307(*)	-0.052	0.152	.332(*)	-0.054	0.268	-0.074	-0.038	-0.301	0.142	0.007
D				0.177	-0.045	0.141	0.091	-0.131	0.081	-0.055	-0.042	-0.08	0.131	-0.003
E					.451(**)	.638(**)	.613(**)	0.188	.800(**)	.589(**)	0.219	0.12	.470(**)	0.221
F						.699(**)	.764(**)	-0.09	.428(**)	.508(**)	0.086	.430(**)	0.282	0.062
G							.753(**)	-0.161	.667(**)	.619(**)	0.1	.325(*)	0.302	0.27
H								0.077	.656(**)	.552(**)	0.027	0.247	.359(*)	0.197
I									0.247	0.264	0.183	0.116	0.118	0.026
J										.719(**)	0.173	0.124	.524(**)	0.271
K											.456(**)	.730(**)	.649(**)	.431(**)
L												.446(**)	.480(**)	0.082
M													.515(**)	.306(*)
N														0.222

*Correlation significance at 0.05

** Correlation significance at 0.01

Key:

A	B	C	D	E	F	G	H	I	J	K	L	M	N	O
Level of true branching	Height at first apical branching	Level of lateral branching	Height at first lateral branching	Plant height	Central lobe length	Central lobe width	Petiole length	Number of rotten tuber	Shoot weight	Root weight per plant	Starch yield (%)	Harvest index (%)	Root length	Root diameter

Table 9: Principal component analysis showing the contribution of morphological traits among the cassava genotypes

Variables	PC1	PC2	PC3
Colour of young leaf	0.045	-0.412	0.073
Pubescences on young leaf	-0.003	-0.133	-0.289
Leaf vein colour	0.071	-0.867	-0.116
Central lobe length	0.85	0.063	0.319
Petiole length	0.194	-0.042	0.062
Stem colour	-0.025	0.112	-0.204
Root length	0.183	-0.043	-0.373
Root diameter	0.026	0.135	-0.589
Root surface colour	-0.023	0.095	-0.178
Root pulp colour	0.016	0.027	-0.021
Colour of root cortex	0.339	0.081	-0.192
Petiole colour	0.78	0.069	0.056
Plant height	0.03	-0.022	-0.011
Yield/plant	0.272	0.037	-0.437
Eigenvalue	26.002	8.21	6.352
% Variance	46.6	14.7	11.4
Cumulative variance	46.6	61.3	72.9

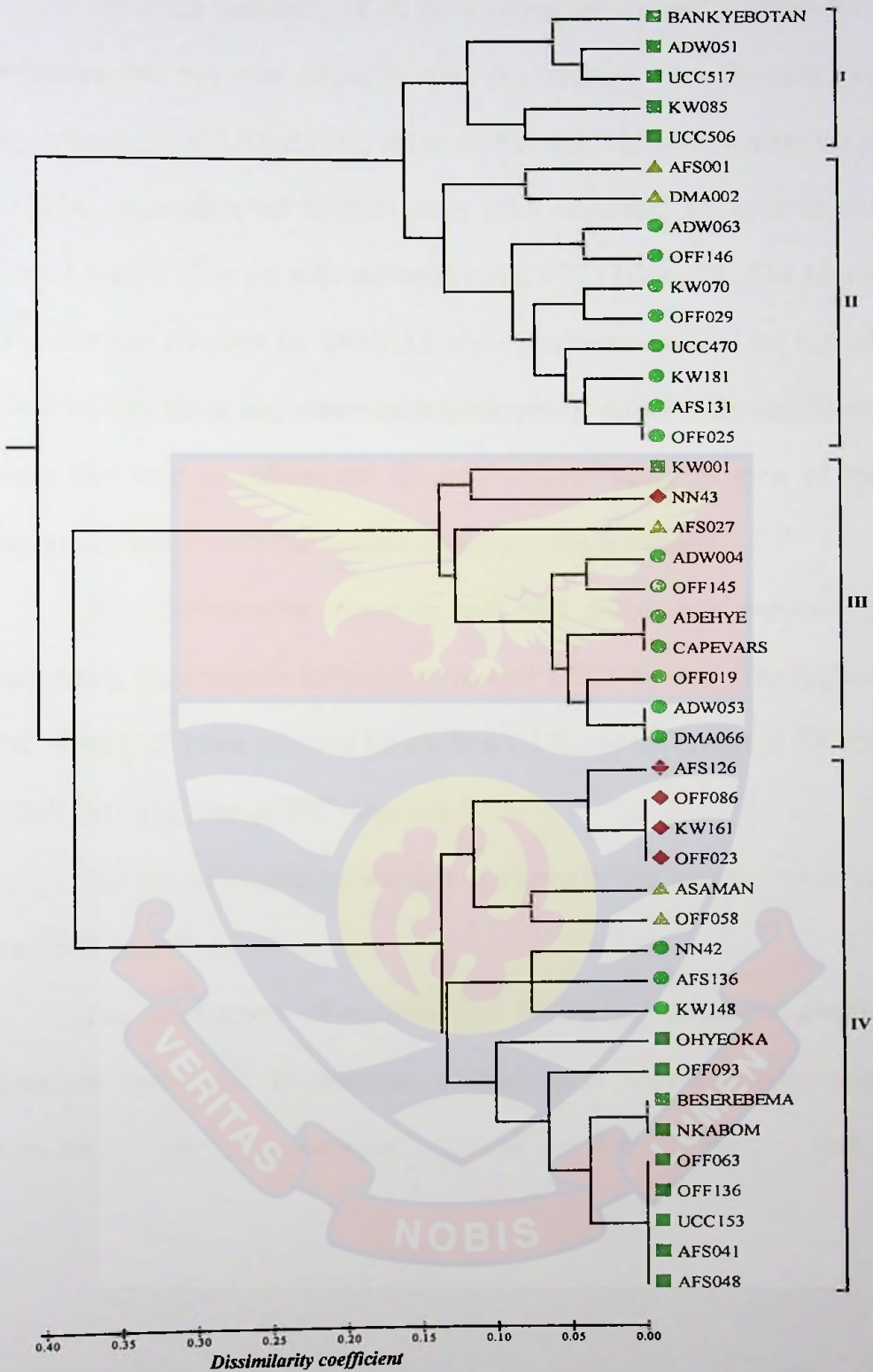


Figure 12: Dendrogram of cassava accessions based on morphological data

The allele frequency of all the primers was generally below 0.95 indicating that they were all polymorphic in character. Gene diversity was high ranging from 0.07 in SSRY181 to 0.78 in SSRY175 with a mean value of 0.58. The observed heterozygosity (H_o) calculated for each primer ranged from 0.07 to 1.0 with the mean being 0.77 (Table 10). The lowest H_o value was recorded for SSRY181 while 8 primers recorded the highest value of 1.0. These high observed heterozygosity values were significant, since they tend to substantiate the heterozygote nature of most of the accessions and the fact that cassava is largely cross pollinated.

The discriminative power of each SSR primer was assessed by calculating polymorphic information content (PIC) values. The highest PIC value (0.75) was recorded for the SSRY 175 and the lowest of 0.07 by SSRY181. The average PIC value was 0.52.

The rate of inbreeding was low as shown by the negative values of the coefficient (Table 10).

The band scores of microsatellite alleles and calculated genetic distances were used to generate a dendrogram showing relationship between the accessions used for the study.

Table 10: Summary of genetic information generated by 20 SSR primers on 43 accessions of cassava

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SSR marker	Allele size Range (bp)	Number of Allele	Allele Frequency	Gene Diversity	Heterozygosity (Ho)	PIC	f
SSRY59	150 – 170	5	0.59	0.59	0.51	0.54	0.14
SSRY20	130 – 160	9	0.37	0.73	0.98	0.68	-0.34
SSRY 21	160 – 230	8	0.73	0.45	0.28	0.44	0.39
SSRY51	240 – 320	5	0.50	0.63	1.00	0.57	-0.58
SSRY64	198 – 250	5	0.47	0.62	1.00	0.55	-0.61
SSRY100	195 – 230	6	0.48	0.67	0.58	0.61	0.14
SSRY101	200 – 250	7	0.36	0.76	1.00	0.72	-0.31
SSRY103	255 – 350	8	0.40	0.72	1.00	0.68	-0.37
SSRY182	220 – 255	7	0.45	0.64	1.00	0.57	-0.56
SSRY175	75 – 110	9	0.35	0.78	0.91	0.75	-0.15
SSRY4	280 – 300	3	0.60	0.50	0.42	0.39	0.17
SSRY19	200 – 250	4	0.45	0.65	1.00	0.58	-0.53
SSRY9	270 – 290	4	0.42	0.69	1.00	0.63	-0.44
SSRY47	249 – 295	3	0.60	0.55	0.79	0.48	-0.44
SSRY69	228 – 239	3	0.76	0.40	0.49	0.36	-0.22
SSRY151	190 – 210	3	0.58	0.54	0.67	0.46	-0.23
SSRY155	145 – 200	3	0.69	0.44	0.60	0.35	-0.37
SSRY179	195 – 226	3	0.42	0.62	1.00	0.54	-0.60
SSRY181	165 – 195	2	0.97	0.07	0.07	0.07	-0.02
NS911	120 – 140	3	0.48	0.63	0.95	0.56	-0.50
Mean		5.0	0.53	0.58	0.78	0.53	-0.27

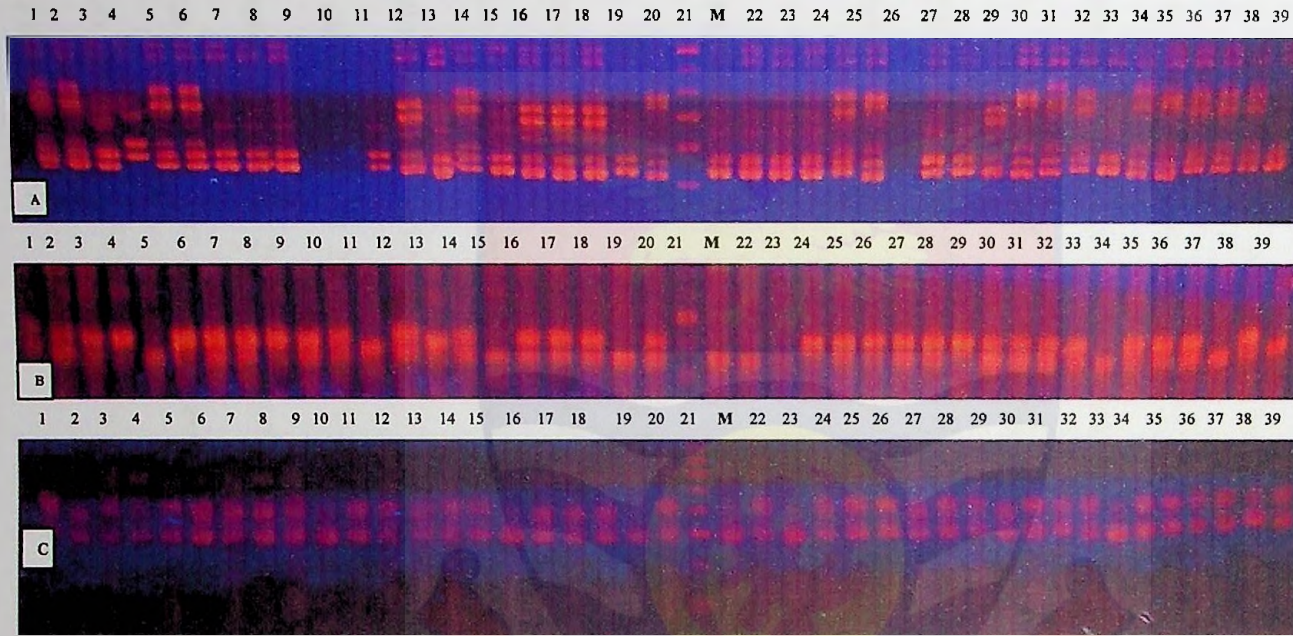


Plate 11: SSR markers profile of cassava accessions generated by primers: A) SSRY9, B) SSRY181 and C) SSRY64.

Using Power Marker version 3.25 (Liu & Muse 2005) software and frequency based distance after Nei and colleagues (1983) a UPGM tree based on number of differences was obtained (Figure. 13). The similarity coefficients ranged from 0.0 to 0.25. The dendrogram revealed 9 distinct subclusters within two main clusters at 0.15 similarity coefficient (Figure. 14). Only two accessions, AFS027 and OFF058 formed one of the main clusters, while the rest were in the other main clusters. The larger cluster was subdivided into subclusters I to VII. The subcluster size varied from 1 accession to 16 accessions. Cluster I, VIII, and IX were made up of only one accession each. Clusters III, IV and V had 2 accessions each. The largest group, cluster VII, had 16 genotypes, and followed by cluster VI with 13 accessions.

Whitefly population

Six weeks after planting (WAP), the overall mean adult whitefly population was 9.7, with a wider range from 1.8 to 28.4 in 2007. More than 50% of the cassava genotypes recorded values below the overall mean value in 2007. However, in 2008 at 6 WAP, the overall mean was 93.2 with a range from 25.4 to 209.9. The mean in 2008 is almost 10 times higher than that of 2007. Capevars recorded the highest mean count of 28.4 and 209.9 for 2007 and 2008 respectively (Table 11). The lowest count was recorded on OFF 086 with a mean value of 1.8 and 25.4 for 2007 and 2008 respectively.

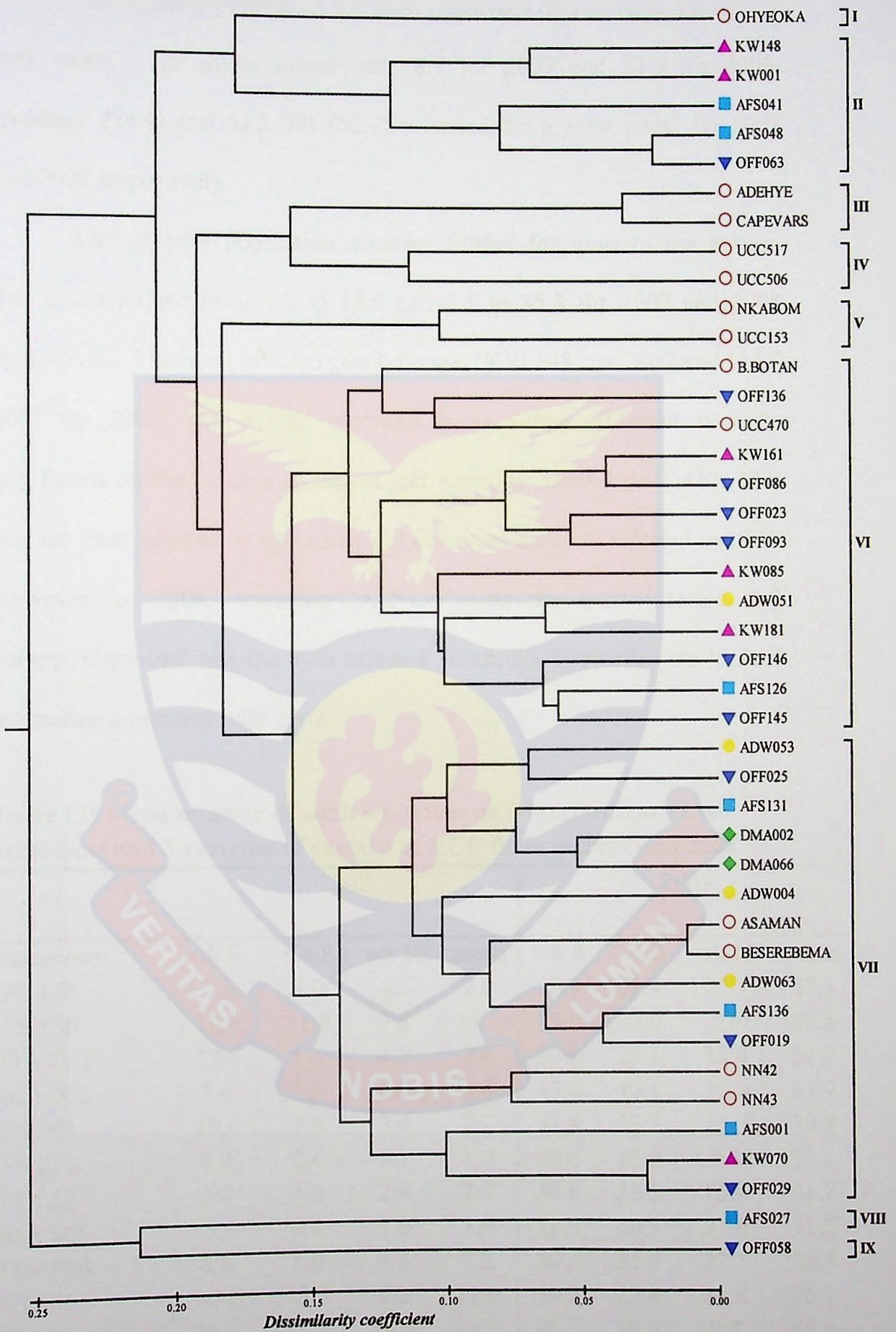


Figure 13: Dendrogram of cassava accessions based on SSR data

The whitefly population for most genotypes decreased at 8WAP for both years. The mean values were 8.7 for 2007 and 33.9 for 2008. 'Adehye' (24.4) and AFS 001 (52.7) recorded the highest count for 2007 and 2008 respectively.

The whitefly population decrease further for most of the entries. The count ranged from 1.1 to 18.6 and 5.5 to 35.3 for 2007 and 2008 respectively. The most infested genotype was 'KW 148' for 2007 and 'AFS 001' for 2008. The overall recorded mean values of adult whitefly population on the cassava accession and varieties showed that 'AFS 027' was the least infested by whiteflies and Capevars the most infested in 2007. However, in 2008, accession 'AFS136' was the least infested and variety 'Capevars' was the most infested. In all, 2008 recorded the highest infestation compared with 2007.

Table 11: Mean number of adult whiteflies on 5 leaves/plant for 40 accessions and 3 varieties of cassava at UCC farm in 2007 and 2008

	2007				2008			
	wk 6	wk 8	wk 10	mean	wk 6	wk 8	wk 10	mean
Accessions								
OFF 146	7.6	9.0	4.5	7.0	53.9	31.4	26.1	37.1
AFS 136	18.0	11.4	3.8	11.1	28.0	29.0	5.6	20.8
ADW 063	12.6	5.6	4.7	7.6	55.0	25.1	15.8	32.0
DMA 002	3.0	5.6	5.7	4.8	43.0	46.1	12.5	33.9
AFS 001	10.6	7.6	3.4	7.2	31.4	52.7	35.3	39.8
AFS 027	2.2	2.4	1.1	1.9	49.6	33.0	9.3	30.6
OFF 058	3.4	3.0	2.4	2.9	56.6	23.2	15.4	31.7
DMA 066	11.6	8.4	3.8	7.9	79.9	46.9	27.0	51.2
ADW 004	8.8	8.0	4.8	7.2	49.6	31.9	27.5	36.3
AFS 131	16.2	13.3	5.2	11.6	39.2	27.4	11.8	26.1
KW 148	25.2	15.3	18.6	19.7	98.7	38.5	9.7	48.9
KW 181	26.4	10.8	7.5	14.9	51.7	42.2	25.8	39.9
ADW 051	13.6	8.2	8.6	10.1	70.3	33.6	13.6	39.2
KW 001	28.4	17.2	16.9	20.8	90.3	44.5	25.7	53.5
OFF 029	5.2	6.0	4.9	5.4	90.0	28.4	8.6	42.3

Table 11 continued

ADW 053	6.2	5.6	4.5	5.4	97.6	33.3	11.8	47.6
KW 085	20.4	16.6	14.0	17.0	71.7	42.9	12.3	42.3
OFF 086	1.8	3.6	3.4	2.9	25.4	30.0	27.2	27.5
OFF 145	10.0	8.8	6.3	8.4	81.8	25.4	11.7	39.6
KW 161	6.5	8.6	7.5	7.5	106.8	31.7	24.4	54.3
OFF 025	5.8	8.2	4.9	6.3	47.0	27.1	12.3	28.8
OFF 023	10.3	10.8	7.8	9.6	127.5	26.5	12.4	55.4
OFF 063	14.0	13.4	14.0	13.8	89.7	45.5	5.5	46.9
AFS 048	14.2	16.6	15.6	15.5	163.8	35.9	15.0	71.6
KW 070	3.4	4.6	3.9	4.0	55.6	21.3	15.0	30.6
AFS 041	11.2	7.6	12.9	10.6	162.8	33.5	10.0	68.8
OFF 093	2.6	2.2	3.3	2.7	134.9	31.6	24.1	63.5
OFF 019	6.6	7.6	4.7	6.3	143.7	34.3	10.1	62.7
AFS 126	4.6	8.6	5.5	6.2	105.6	41.5	14.6	53.9
OFF 136	8.0	8.8	9.6	8.8	165.7	32.6	11.9	70.1
NKABOM*	5.8	8.2	8.2	7.4	175.9	32.7	12.1	73.5
UCC 517	3.0	1.0	2.2	2.1	128.2	35.1	9.1	57.5
UCC506	10.2	6.0	9.7	8.6	161.5	22.4	14.7	66.2
B. BOTAN*	11.6	16.2	8.9	12.2	162.5	26.5	17.0	68.6
ADEHYE	16.8	24.4	16.1	19.1	140.3	38.4	19.6	66.1
CAPEVARs*	26.6	21.6	16.0	21.4	209.9	33.9	26.7	90.2
UCC 470	7.4	1.4	6.2	5.0	93.6	43.9	27.0	54.8
UCC 153	2.2	7.0	10.8	6.7	97.9	30.1	26.2	51.4
BESEREBEMA	3.6	1.4	4.9	3.3	70.9	34.9	10.5	38.7
ASAMAN	2.6	5.8	5.2	4.5	76.6	35.4	16.9	42.9
OHYEOKA	2.0	6.9	6.2	5.0	84.6	30.2	11.1	42.0
NN 42	4.3	4.0	10.2	6.2	56.8	30.5	15.5	34.3
NN43	4.6	8.2	7.0	6.6	85.1	36.0	12.1	44.4
Mean	9.7	8.7	7.6	8.7	93.2	33.9	16.4	47.8
Range	1.8 -	1.0 -	1.1 -	1.9 -	25.4-	21.3-	5.5 -	20.8-
	28.4	24.4	18.6	21.4	209.9	52.7	35.3	90.2
% CV	75.3	60.9	59.2	59.8	49.5	33.9	44.5	32.4

* Released varieties

Cassava mosaic severity scores

Table 12 show the ordinal scores of African cassava mosaic virus disease infection for 2007 and 2008 growing seasons. At 6 week after planting (WAP) in 2007 the mean score for all the cassava genotypes on the

field was 2.8, with a range score of 1-5. With this range of scores, 5 genotypes had a score of 1, 12 genotypes had a score of 2, 14 genotypes recorded a score of 3, 9 genotypes scored 4 and 3 genotypes registered the high score of 5. DMA 002, ADW 004 and OFF 029 were the most susceptible to ACMV disease at that growth stage of the plant with the highest approximate mean score of 5.

On the 12 WAP, 4 genotypes gave a score of 1, 12 genotypes for score of 2, 16 gave a score of 3, 9 recorded score of 4 and 3 registered scores of 5. The mean severity score was 2.9 for 2007. At the same age in 2008 the distribution of mosaic were; 4, 9, 7, 20, and 4 genotypes for scores of 1, 2, 3, 4, and 5 respectively. The mean score for 2008 was 3.3. Comparatively the mean score shows that more cassava genotypes were more susceptible to ACMV in 2008 than in 2007.

Table 12: CMD severity on 40 accessions and 3 varieties for 2007 and 2008 cropping seasons

Accession	2007				2008			
	wk 6	wk 12	wk 20	wk 48	wk 6	wk 12	wk 20	wk 48
OFF 146	3.7	4.2	3.1	2.1	5.0	4.2	3.7	3.1
AFS 136	3.0	2.7	2.2	1.9	4.7	3.7	3.1	1.4
ADW 063	4.0	3.1	3.2	1.2	5.0	4.4	2.8	1.2
DMA 002	4.7	5.0	4.0	1.0	5.0	4.1	4.1	1.3
AFS 001	4.0	3.8	2.7	2.8	4.2	4.0	5.0	4.3
AFS 027	3.1	4.1	3.0	2.2	3.1	4.4	4.3	1.0
OFF 058	4.3	2.7	2.8	2.1	4.0	3.2	4.1	3.1
DMA 066	3.1	4.1	3.1	1.3	4.1	3.0	3.3	1.0
ADW 004	4.6	5.0	4.1	4.0	5.0	4.1	4.4	1.2
AFS 131	4.4	4.3	3.2	3.2	5.0	4.0	3.6	1.1
KW 148	2.1	3.1	2.0	1.0	3.8	3.2	2.8	1.2
KW 181	3.3	3.3	3.0	2.1	4.7	4.8	4.2	1.4
ADW 051	3.1	3.1	2.1	1.0	3.3	2.4	3.1	2.4
KW 001	1.5	2.8	1.8	1.0	4.0	2.1	3.3	1.1
OFF 029	4.6	4.8	3.5	1.8	5.0	5.0	5.0	3.2
ADW 053	2.9	3.1	1.6	1.0	3.1	2.3	4.3	1.3

Table 12 continued

KW 085	1.0	1.0	1.0	1.0	1.0	1.0	2.2	1.0
OFF 086	3.1	2.6	3.0	2.0	4.7	3.4	3.1	2.2
OFF 145	2.2	3.3	4.0	3.7	4.0	4.0	5.0	4.1
KW 161	3.1	2.4	3.1	1.0	4.2	3.1	4.1	2.0
OFF 025	1.8	3.9	4.3	2.0	5.0	4.3	4.8	3.2
OFF 023	2.8	2.6	2.0	2.0	3.1	4.0	1.7	1.3
OFF 063	1.0	1.7	2.1	1.0	2.3	2.0	1.8	1.2
AFS 048	2.1	2.1	1.7	1.0	2.1	2.3	2.1	2.3
KW 070	3.8	3.0	5.0	1.0	4.3	4.6	4.8	1.0
AFS 041	2.0	2.1	2.0	2.7	1.5	2.4	2.2	2.1
OFF 093	3.0	3.0	2.8	1.0	3.2	4.3	4.2	1.0
OFF 019	2.3	2.0	2.1	1.0	3.3	2.8	2.4	2.0
AFS 126	4.1	3.7	3.9	1.0	5.0	4.1	5.0	4.4
OFF 136	2.1	3.0	2.1	1.7	2.2	2.1	2.1	2.0
NKABOM*	2.4	2.3	1.5	1.1	1.9	1.9	2.0	2.0
UCC 517	2.7	2.1	2.0	1.8	3.4	4.1	3.2	2.0
UCC506	2.2	2.0	1.3	1.1	1.6	3.2	4.1	1.3
B. BOTAN*	1.0	1.0	2.0	1.0	1.6	3.5	2.6	2.4
ADEHYE	1.0	1.0	1.2	1.0	1.0	1.0	1.0	1.0
CAPEVARs*	1.0	1.0	1.1	1.0	1.0	1.0	1.0	1.0
UCC 470	2.0	2.2	2.0	1.2	2.2	1.4	2.0	2.0
UCC 153	1.8	2.0	3.0	1.3	3.0	2.2	2.3	2.0
BESEREBEMA	3.1	2.8	1.8	1.1	4.4	3.8	4.7	1.3
ASAMAN	4.0	3.6	3.0	3.0	5.0	4.8	5.0	3.3
OHYEOKA	3.0	4.0	4.0	3.1	5.0	3.7	4.0	1.1
NN 42	2.9	2.0	3.0	2.2	4.2	4.4	3.6	1.4
NN43	4.0	3.1	3.5	3.8	5.0	4.1	5.0	1.0
Mean	2.8	2.9	2.6	1.7	3.5	3.3	3.4	1.9
Range	1 - 5	1 - 5	1 - 5	1 - 4	1 - 5	1 - 5	1 - 5	1 - 4
%CV	39.3	35.9	38.5	52.9	37.1	33.3	35.3	52.6

*Released varieties

At 20 WAP in 2007 the mean score was 2.6 and that of 2008 was 3.4 with both years recording a range scores from 1- 5. Again, 2008 season recorded the highest mean severity score which depicts that more cassava genotypes were susceptible in 2008 than in 2007.

At 48 WAP, which was the harvest time, ACMD severity score was recorded to have an idea about how the genotypes have recovered from the

disease. The average mean scores were 1.7 and 1.9 for 2007 and 2008 respectively. However, in both years 23 genotypes recorded score of 1, 12 genotypes gave a score of 2, 5 genotypes recorded a score of 3 and 3 genotypes recorded a score of 4. None of the cassava genotype recorded the most severity score of 5.

The mean CMD responses scores for all the 43 cassava genotypes, by pooling the 6WAP, 12WAP and 20WAP for the two years together, revealed varying levels of resistance and susceptibility (Figure 14). Three genotypes were classified as highly resistant (HR) with a score of 1, ten as resistant (R) with a score of 2, thirteen as susceptible (S) and seventeen as highly susceptible (HS) with scores of 4 and 5.

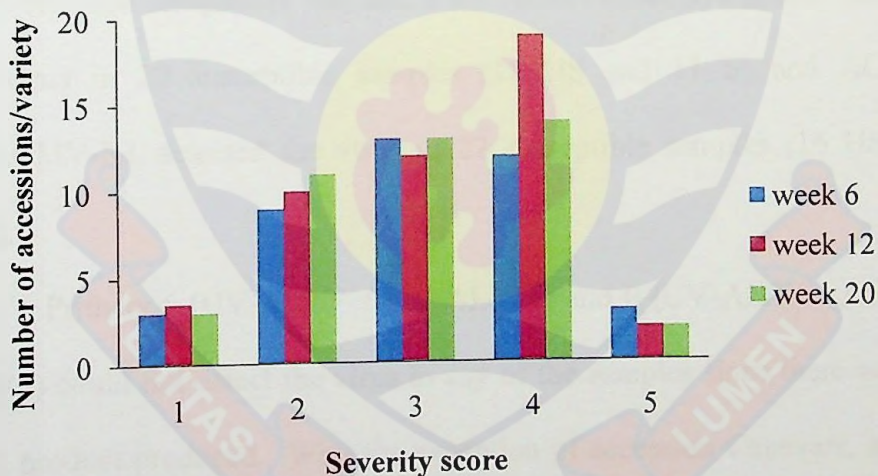


Figure 14: Mean CMD severity of pooled data for 2007 and 2008 planting seasons

Mosaic screening at the laboratory

Six pairs of microsatellite primers that detect three strains of cassava mosaic viruses as described earlier in Table 6 were used. The PCR diagnostics test detected only ACMV as the causal agent responsible for the characteristic symptoms in the population. The ACMV specific primers that were more reliable in detecting the virus were ACMV-1/ACMV-2 and ACMVF1/ACMV-R1. The ACMV specific primers ACMV-1/ACMV-1 and ACMV-F1/ACMV-R1 detected ACMV in 39 and 38 accessions, respectively.

ACMV-1/ACMV-2 detected the virus in 10 resistant samples (2 HR and 8 R samples) while ACMV-F1/ACMV/R1 also detected the virus in 10 resistant samples (1 HR and 9 R). ACMV-1/ACMV-2 also detected the virus in 29 susceptible samples (18 HS and 11 S) and ACMV-F1/ACMV-R1 detected the virus in 27 susceptible samples (16 HS and 11S).

Primers 5 (UV-AL3/F + UV-ALI/F1) and 6 (UV-AL/F1 + ACMV-CP/R3) could not detect the virus in any of the samples since there was no PCR product produced. With the exception of accession Capevars, all the samples were susceptible to one or more of the viruses. However, Nkabom and Adehye were susceptible to ACMV1 and ACMV3, respectively (Table 12). ACMV-ALI/FACMV-ARO/R recorded the highest PIC value which shows that it was more polymorphic in detecting ACMV. ACMV-F1/ACMV-R1 and ACMV-1/ACMV-2 recorded the same PIC value of 0.18 (Table 13).

The forty three cassava genotypes were clustered into 11 groups at a similarity coefficient of 0.13 based on the PCR amplification products. The cluster size ranged from 1 to 23. Clusters 1, 2, 3, 6 and 8 had one genotype each. Cluster 11 had the highest number of accessions (Figure. 15).

Resistance Screening

From the results obtained from PCR reactions with ACMV specific primers (Plate 13) and field screening for mosaic resistance, four accessions were selected for further screening with markers associated with CMD2 gene that confer resistance to CMD to ascertain their source of resistance.

The four markers used were SSRY28, NS158, NS169 and RME1. The gel profiles for the various markers are shown in Plate 13. All the genotypes selected had bands for alleles of the genes in all the four markers. However, the bands present were brighter in two markers (NS169 and RME1).

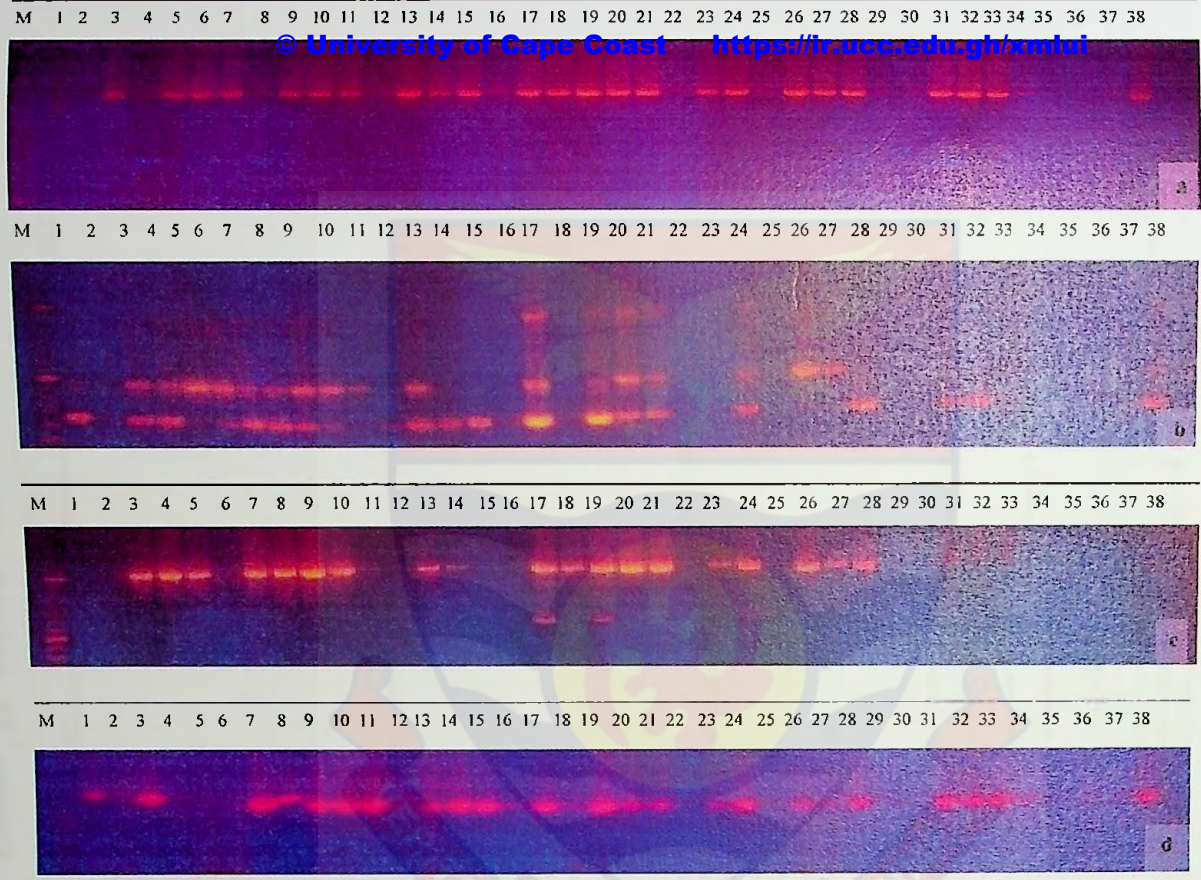


Plate 12: Gel profile for ACMV specific primers: a) ACMV-F1/ACMV-R1, b) ACMV-F2/ACMV-R2, c) ACMV-AL1/F/ACMV-ARO/R and d) ACMV-1/ACMV-2

Table 13: Gene diversity and PIC values for ACMV primers

Primer	Gene Diversity	PIC
1. ACMV1	0.21	0.18
2. ACMV2	0.33	0.28
3. ACMV-AL	0.44	0.34
4. ACMV3	0.21	0.18
Mean	0.30	0.25

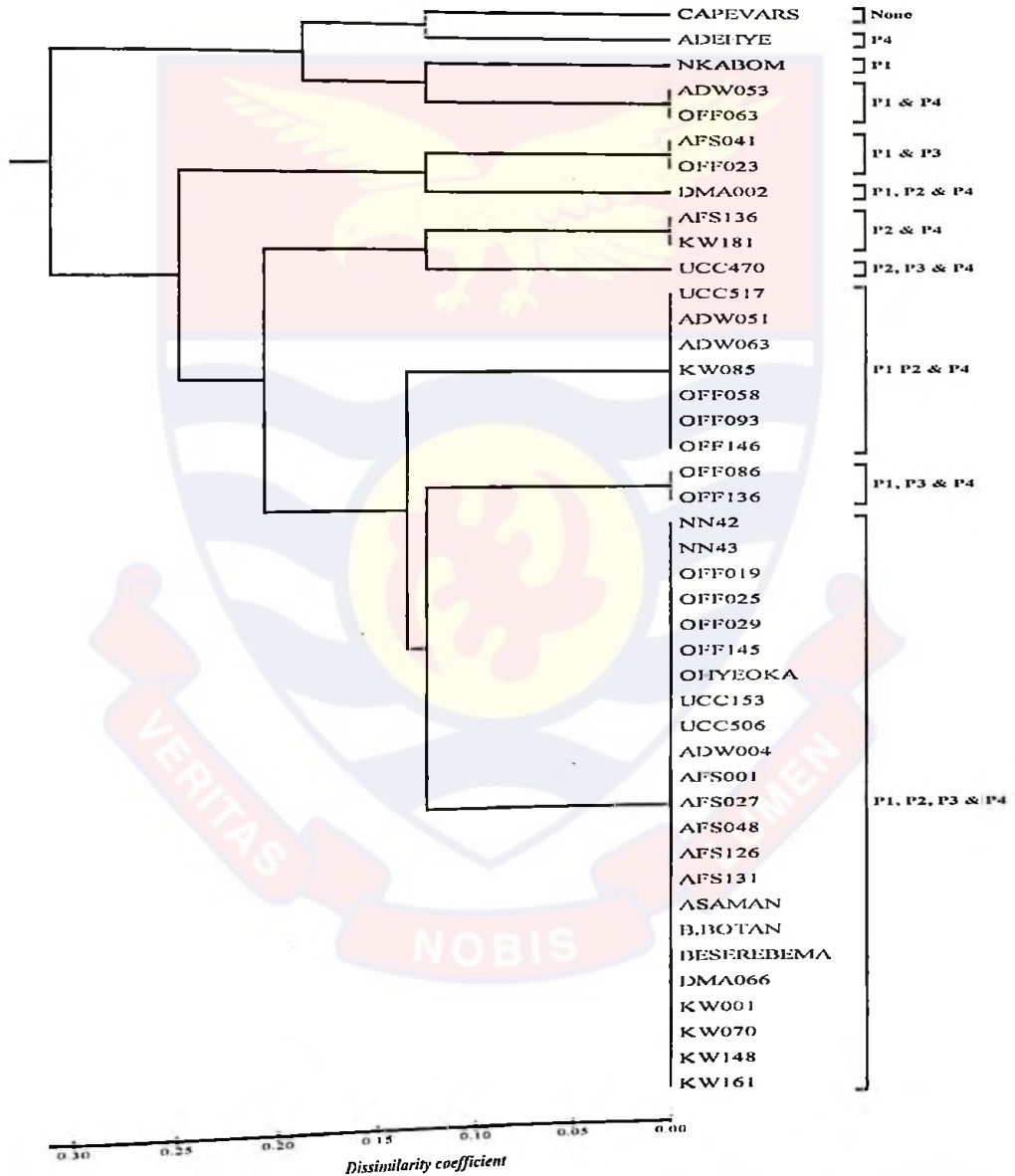


Figure 15: Dendrogram showing 43 cassava accessions reaction patterns with four mosaic primers as determined by the unweighted pair group method with arithmetic averages of binary character matrices using the similarity coefficient index (Nei, 1983)

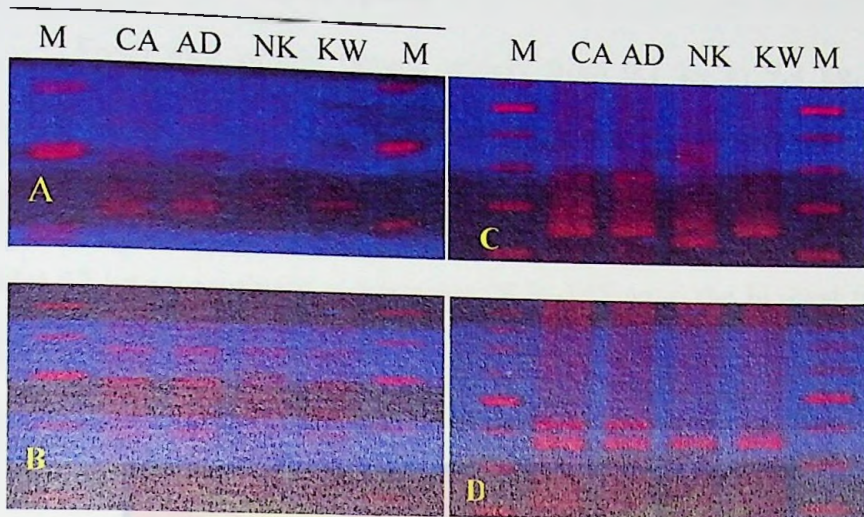


Plate 13: PCR amplifications of 4 cassava genotypes on PAGE gel stained with ethidium bromide, using four markers associated with CMD resistance namely SSRY28 (A), NS158 (B), NS169 (C) and RME1 (D). M- Marker, CA- Capevars, AD- Adehye, Nkabom and KW-KW085.

Agronomic and production traits

Agronomic and production traits of cassava are the traits of cassava recorded at twelve months after planting (MAP). Table 14 summaries the result of most relevant agronomic and production traits.

Number of roots per plant

At twelve months after planting, the average number of roots per plant was 4.1, with a range of variation from 0.3 to 9.4 and percentage coefficient of variation (%CV) of 50%. UCC 517 recorded the highest (9.4) number of roots followed by accession 'AFS 041' with 7.3 roots per plants. Accession 'KW 070' gave the lowest number of roots of 0.3. Fifty percent of the cassava genotype recorded values higher than the mean.

Fresh root weight (kg) per plant

Fresh root weight ranged widely from 0.1 kg to 9.4 kg, with an average of 3.1 kg and %CV of 66.1. Capevars, Nkabom and Bankye Botan which are released varieties among the genotypes recorded 3.51 kg, 3.87 kg and 4.57 kg respectively. Accessions 'KW 148' gave the highest fresh root weight (9.38 kg) and 'KW 070' and 'OFF 029' recorded the lowest fresh root weight of 0.10 kg. Nine accessions recorded values less than 1 kg.

Mean root weight (g)

The mean root weight per tuber ranged from 133.3 g to 1,465.6 g for accessions 'KW 148' and 'OFF 093' respectively. The average mean root weight was 700.9 g. Only 5 cassava genotype out of 43 recorded values above 1000 g.

Fresh shoot weight (kg/plant)

Fresh shoot weight of the cassava genotypes are presented in Table 14. Accession 'AFS 041' registered the highest fresh shoot weight of 14.84 kg followed by 'KW 148' with 13.05 kg. Accession 'KW 070' recorded the lowest value of 0.95 kg. The mean fresh shoot weight was 4.84 kg and percent coefficient of variation of 62 %. Three accessions recorded values above 10 kg.

Fresh root yield (t ha⁻¹)

The fresh root yield (t ha⁻¹) ranged widely from 1.0 t/ha to 93.8 t/ha, with an average of 31.02 t ha⁻¹. Accession 'KW148' out yielded all the

other genotype with yield value of 93.80 t ha⁻¹. The closest competitor was 'KW001' with a yield value of 62.50 t ha⁻¹. Accessions 'OFF 029' and 'KW 070' recorded the lowest yield of 1.00 t ha⁻¹. The three released varieties gave yield value of 35.10 t ha⁻¹, 38.70 t ha⁻¹ and 45.70 t ha⁻¹ for Capevars, Nkabom and Bankye Botan respectively.

Harvest index (%)

The percentage harvest index at 12 MAP for the cassava genotypes ranged from the lowest value of 3.09 % to highest value of 58.6 % for accessions 'KW 085' and 'Adehye' respectively. The average percentage harvest index was 36.43 %. The values recorded by the three varieties were 44.59 %, 46.49 % and 54.44 % for Nkabom, Capevars and Bankye Botan respectively.

Starch Data

Starch content (%)

Percentage starch values for the 43 cassava genotypes are presented in Table15. The average percentage starch content was 19.4 % and ranged from 14.64 % to 24.83 %. Accession 'AFS 048' gave the highest value of 24.83 % closely followed by accession 'ADW 053' with a value of 24.1 %. Accession 'NN 43' registered the lowest value of 14.64 %.

Starch yield (t ha⁻¹)

At 12MAP the average starch yield recorded by 43 cassava genotype planted at UCC farms was 6.31 t/ha, with a wide range of 0.17 t

ha⁻¹ to 21.89 t/ha. Accessions 'KW 148', 'KW001', 'ADW 053' and variety 'Bankye Botan' were the four top starch yielders. Varieties 'Capevars (6.7 t ha⁻¹) and Nkabom (8.4 t ha⁻¹) performed poorly compared to the highest starch yielder.

Functional properties of starches

Swelling volume values ranged from the lowest value of 14.5 mL g⁻¹ indicated by starch from 'AFS 136' to the highest of 35.3 mL g⁻¹ registered by starch from OFF 093. The mean swelling volume was 29.2 mL g⁻¹ (Table 16).

Accession 'KW 181' produced starch that had the lowest solubility of 8.3 % and 'OFF 146' the starch with the highest value of 27.5 %. The range was quite narrow. The mean solubility value was 14.0 % and more than 50 % of the genotypes recorded values below this mean value.

ADW 063 starch produced the highest swelling power values of 41.6 g g⁻¹. Accession 'KW 001' starch registered the lowest swelling power values of 16.0 g g⁻¹. The average swelling power value was 34.1 g g⁻¹. All the released varieties recorded values above the average value

Table 14: Agronomic and production trait of 42 cassava accession and one variety at UCC farm

Accessions/ Varieties	Number of roots /plant	Fresh root wt (kg)/plant	Mean root weight (g)	Fresh shoot weight (kg/plant)	Fresh root yield (t ha ⁻¹)	Harvest Index (%)
OFF 146	3.1	1.65	532.3	5.05	16.50	24.63
AFS 136	5.8	4.39	756.9	6.43	43.90	40.59
ADW 063	1.5	0.95	633.3	3.05	9.50	23.75
DMA 002	2.9	2.20	758.6	3.35	22.00	39.64
AFS 001	4.0	4.61	1152.5	6.39	46.10	41.91
AFS 027	2.8	1.55	553.6	1.40	15.50	52.54
OFF 058	3.6	2.90	805.6	5.60	29.00	34.12
DMA 066	4.9	2.81	573.5	4.45	28.10	38.71
ADW 004	1.9	0.67	341.0	2.15	6.65	23.62
AFS 131	3.8	4.15	1092.1	4.60	41.50	47.43
KW 148	6.4	9.38	1465.6	13.05	93.80	41.82
KW 181	3.6	1.34	370.8	2.49	13.35	34.95
ADW 051	4.9	5.12	1044.9	3.80	51.20	57.40
KW 001	4.6	6.25	1358.7	9.26	62.50	40.30
OFF 029	0.6	0.10	166.7	1.75	1.00	5.41
ADW 053	6.7	5.29	789.6	4.27	52.90	55.33
KW 085	0.9	0.15	166.7	4.70	1.50	3.09
OFF 086	0.5	0.50	1000.0	2.65	5.00	15.87
OFF 145	5.0	3.85	770.0	3.85	38.50	50.00
KW 161	6.1	3.82	626.2	5.56	38.20	40.72
OFF 025	2.4	0.90	375.0	4.55	9.00	16.51
OFF 023	6.7	3.68	549.3	4.04	36.80	47.67
OFF 063	6.0	5.90	983.3	8.11	59.00	42.11
AFS 048	3.7	2.80	756.8	9.10	28.00	23.53
KW 070	0.3	0.10	333.3	0.95	1.00	9.52

Table 14 continued

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AFS 041	7.3	5.86	802.7	14.84	58.60	28.31
OFF 093	1.5	0.20	133.3	1.55	2.00	11.43
OFF 019	5.2	3.80	730.8	4.42	38.00	46.23
AFS 126	1.7	0.80	470.6	3.50	8.00	18.60
OFF 136	4.8	4.80	1000.0	11.83	48.00	28.86
NKABOM*	5.6	3.87	691.1	4.81	38.70	44.59
UCC 517	9.4	2.90	308.5	2.50	29.00	53.70
UCC506	6.5	4.20	646.2	5.00	42.00	45.65
B. BOTAN*	6.0	4.57	761.7	3.74	45.70	54.99
ADEHYE	5.4	4.60	851.9	3.25	46.00	58.60
CAPEVARs*	4.1	3.51	856.1	4.04	35.10	46.49
UCC 470	4.2	2.90	690.5	5.45	29.00	34.73
UCC 153	6.0	5.92	986.7	7.00	59.20	45.82
BESEREBEMA	3.5	1.50	428.6	2.70	15.00	35.71
ASAMAN	3.3	2.10	636.4	4.30	21.00	32.81
OHYEOKA	2.5	2.00	800.0	2.55	20.00	43.96
NN 42	2.4	1.25	520.8	2.40	12.50	34.25
NN43	4.1	3.55	865.9	3.45	35.50	50.71
Mean	4.1± 0.3	3.1± 0.3	700.9± 45.6	4.84±0.46	31.02±3.14	36.43±2.21
Range	0.3 - 9.4	0.1 - 9.38	133.3 - 1465.6	0.95 - 14.84	1.00 - 93.8	3.09 - 58.60
%CV	50.0	66.1	32.7	62.0	66.3	39.8
Lsd	2.422	1.93	492.2	4.40	19.28	15.94

* Released varieties

Table 15: Starch data of 40 cassava accessions and 3 varieties at twelve months after planting at UCC farms.

Accessions	Starch content (%)	Starch yield (t ha ⁻¹)
OFF 146	15.0±1.4	2.5±1.3
AFS 136	22.0±1.2	9.7±2.1
ADW 063	15.0±0.8	1.4±0.8
DMA 002	19.0±2.1	4.2±1.2
AFS 001	20.6±1.3	9.5±3.7
AFS 027	21.0±1.8	3.3±0.6
OFF 058	20.8±1.0	6.0±1.2
DMA 066	19.6±0.5	5.5±3.2
ADW 004	19.4±0.6	1.3±0.2
AFS 131	20.9±2.5	8.7±3.3
KW 148	23.3±1.2	21.9±8.2
KW 181	15.7±1.2	2.1±1.8
ADW 051	19.0±0.2	9.7±3.4
KW 001	23.8±0.8	14.9±0.8
OFF 029	17.2±0.7	0.2±0.3
ADW 053	24.1±0.5	12.8±2.4
KW 085	19.0±1.2	0.3±1.3
OFF 086	15.1±0.3	0.8±1.1
OFF 145	22.5±0.5	8.7±0.4
KW 161	18.7±0.8	7.2±1.9
OFF 025	16.1±2.6	1.5±0.7
OFF 023	17.4±1.7	6.4±0.8
OFF 063	16.4±1.7	9.7±2.4
AFS 048	24.8±0.2	7.0±1.3
KW 070	17.2±1.2	0.2±0.6
AFS 041	21.8±1.4	12.8±3.2
OFF 093	18.3±0.1	0.4±0.1
OFF 019	23.0±0.7	8.7±2.1
AFS 126	17.4±0.2	1.4±0.6
OFF 136	20.6±0.7	9.9±4.1
NKABOM*	21.7±0.3	8.4±2.7
UCC 517	22.2±1.1	6.4±1.9
UCC506	16.6±1.4	7.0±0.9
B. BOTAN*	22.0±0.7	10.0±3.3
ADEHYE	21.0±0.5	9.6±2.1
CAPEVARs*	19.1±1.1	6.7±1.5
UCC 470	24.3±1.2	7.1±1.8
UCC 153	17.2±2.6	10.2±4.6
BESEREBEMA	20.5±0.5	3.1±1.3
ASAMAN	19.1±0.9	4.0±0.6
OHYEOKA	17.3±2.1	3.5±0.5
NN 42	15.8±2.5	2.0±0.2
NN43	14.6±2.6	5.2±0.8

Table 15 continued

Mean	19.4±0.4	6.31±0.7
Range	14.64 - 24.83	0.17 - 21.89
%CV	14.71	72.74
Lsd	6.87	5.04

* Released variety

Table 16: Functional properties of starch from 40 cassava accessions and 3 varieties

Accessions/variety	Swelling volume (mL g ⁻¹)	Solubility (%)	Swelling Power(g g ⁻¹)
AFS 136	14.5±2.3	13.7±1.7	16.2±2.8
KW 001	14.6±2.0	9.7±1.8	16.0±2.2
KW 181	32.7±1.3	8.3±0.7	36.2±1.0
OFF 063	29.3±0.3	12.3±0.7	35.2±1.5
SECO	30.7±1.7	12.7±0.9	35.3±1.5
BESEREBEMA	28.0±2.0	14.0±0.6	32.4±2.3
AFS 131	29.0±1.0	10.0±0.6	32.7±0.8
OFF 023	29.7±0.9	8.7±1.2	33.0±0.5
OFF 019	30.7±2.3	10.7±0.7	26±5.0
NN 42	29.0±2.3	11.0±0.6	32.5±1.9
ADEHYE	32.7±0.3	10.3±0.9	36.7±0.5
CAPEVARs*	31.0±1.0	15.0±0.6	37.4±1.0
OFF 134	27.7±0.9	11.0±0.6	31.7±0.5
AFS 048	25.3±1.5	11.3±2.9	28.2±2.1
OFF 146	23.3±0.3	27.5±1.5	31.7±0.7
OFF 058	32.3±1.5	14.3±1.5	38.2±0.8
UCC 506	31.8±1.4	15.0±0.0	37.1±0.9
OFF 145	33.0±1.5	13.3±0.7	38.6±1.6
OFF 029	34.8±0.2	12.3±0.3	41.5±0.9
OFF 025	29.5±0.3	16.0±0.6	34.2±0.6
ADW 051	34.8±0.4	11.3±1.9	40.8±0.6
AFS 027	35.0±0.3	10.3±0.3	40.2±0.2
AFS 041	27.8±0.9	19.0±0.6	34.7±0.7
KW 148	29.0±1.0	14.7±1.2	34.5±0.6
AFS 126	32.5±1.5	13.3±2.0	37.5±0.8
DMA 066	31.0±1.5	15.7±0.7	36.8±1.5
ADW 053	26.3±4.3	15.0±0.6	30.8±5.3
ADW 063	31.8±1.4	15.3±1.9	41.6±3.7
ADW 004	30.5±1.0	14.7±1.2	35.7±0.9
DMA 002	25.0±0.0	19.0±0.6	31.4±0.3
KW 085	30.2±1.2	13.3±1.2	34.7±0.6
OHYEOKA	25.3±0.7	21.3±2.8	33.1±0.3
UCC 470	26.8±0.9	15.7±1.2	32.2±0.5

Table 16 continued

UCC 517	28.2±1.4	16.7±2.3	34.1±0.5
NKABOM*	33.8±0.4	13.3±1.9	39.2±0.1
OFF 093	35.3±1.6	13.3±3.3	40.5±0.4
OFF 086	24.2±0.2	19.0±0.6	30.0±0.1
B. BOTAN*	29.8±0.9	16.3±1.3	35.8±0.7
KW 161	34.7±0.3	11.7±0.3	37.7±1.0
AFS 001	29.0±1.0	12.7±0.7	33.8±1.2
ASAMAN	29.7±0.3	13.0±3.2	34.4±1.2
KW 070	23.4±1.2	11.8±0.8	36.8±1.6
NN 43	26.5±0.7	18.0±0.0	32.4±0.6
Range	14.5 - 35.3	8.3 - 27.5	16 - 41.6
Mean	29.2±0.7	14.0±0.6	34.1±0.8
Lsd	3.88	3.99	4.67

* Released varieties

Table 17 gives the ranks of 40 cassava accessions and 3 varieties in terms of swelling volume, percentage solubility and swelling power. The overall 15 outstanding cassava genotypes in terms of high swelling volume, low solubility and high swelling power were 'AFS027', 'ADW051', 'KW070', 'OFF 029', 'KW 181', 'KW 161', 'OFF 093', 'ADEHYE', 'NKABOM' and 'OFF 145'. The rest were: 'AFS 126', 'OFF058', 'ADW063', 'UCC506' and 'OFF023'.

Table 17: Rankings of 40 cassava accessions and 3 varieties in terms of Swelling Volume, Solubility and Swelling Power

Accessions/ Varieties	Swelling Volume	Solubility	Swelling Power	Total	Final ranking
AFS 027	2	5.5	6	13.5	1
ADW 051	3	10.5	4	17.5	2
KW 070	6	13	1	20	3
OFF 029	4	14.5	3	21.5	4
KW 181	9	1	16	26	5
KW 161	5	12	10	27	6
OFF 093	1	21	5	27	7
ADEHYE	10	5.5	15	30.5	8

Table 17 continued

NKABOM	7	21	7	35	9
OFF 145	8	21	8	37	10
AFS 126	11	21	11	43	11
OFF 058	12	26	9	47	12
ADW 063	14	32	2	48	13
UCC 153	18	16.5	19	53.5	14
OFF 023	23	2	29	54	15
UCC 506	13	30	13	56	16
CAPEVARs	15	30	12	57	17
OFF 063	25	14.5	20	59.5	18
AFS 131	26	4	30	60	19
KW 085	20	21	22	63	20
DMA 066	16	33.5	14	63.5	21
ASAMAN	22	18	24	64	22
ADW 004	19	27.5	18	64.5	23
OFF 019	17	7	41	65	24
NN 42	29	8.5	31	68.5	25
AFS 001	28	16.5	27	71.5	26
B. BOTAN	21	36	17	74	27
OFF 136	33	8.5	35	76.5	28
KW 148	27	27.5	23	77.5	29
OFF 025	24	35	25	84	30
AFS 048	37	10.5	40	87.5	31
KW 001	42	3	43	88	32
BESEREBEMA	31	25	32	88	33
UCC 517	30	37	26	93	34
AFS 041	32	40	21	93	35
UCC 470	34	33.5	34	101.5	36
ADW 053	36	30	38	104	37
NN 43	35	38	33	106	38
OHYEOKA	38	42	28	108	39
AFS 136	43	24	42	109	40
DMA 002	39	40	37	116	41
OFF 086	40	40	39	119	42
OFF 146	41	43	36	120	43

Pasting characteristics of cassava starch

Pasting temperature (°C)

Pasting temperature of cassava starch extracted from 43 cassava genotypes are presented in Table 18. The mean pasting temperature was 65.8 with very narrow range 63.8 to 68.2 °C. Accession 'ADW 053' gave the highest value of 68.2 °C and accession 'OFF 086' recorded the lowest value of 63.8 °C.

Peak temperature (°C)

The peak temperature values recorded from starches from cassava genotypes planted at UCC varied from 67.3 to 73.5 °C from tubers of accessions 'OFF 086' and 'ADW 053' respectively. The mean peak temperature of the starches was 70.7 °C. Among the three varieties Bankye Botan recorded the highest peak temperature of 71.6 °C with Nkabom and Capevars recording 71.1 °C and 69.9 respectively.

Paste stability at 95 °C (BU)

Paste stability at 95 °C values recorded for the different starches ranged from the lowest of 21.5 BU to the highest value of 140.0 BU. Accession 'UCC 506' and variety 'Bankye Botatn recorded the lowest and highest values respectively. The mean value was 87.7 BU. Less than 50 % of the 43 cassava genotype had values greater than 100 BU.

Paste stability at 50 °C (BU)

Paste stability at 50 °C values obtained varied widely for a very low of -28.0 BU to 22.0 BU. Accessions 'AFS048 and 'ADW 063' registered the lowest and highest values respectively. The mean value of 3.5 BU was recorded. Several accessions (30.2 %) recorded negative values.

Table 18: Pasting characteristics of starch from 40 cassava accessions and 3 varieties

Accessions/ Varieties	Pasting Temperature (°C)	Peak Temperature (°C)	Paste stability at 95°C	Paste stability at 50°C
Beserebema	65.4	69.2	77.0	-2.5
KW 148	65.2	71.5	74.5	-17.0
KW 070	66.4	70.9	81.5	-5.5
AFS 048	64.6	70.3	52.5	-28.0
UCC 153	66.1	69.6	50.0	20.5
KW 001	65.9	72.0	82.0	21.5
KW 181	65.9	70.5	115.5	3.0
OFF 029	65.5	70.0	104.5	0.5
OFF 023	65.7	70.5	117.0	0.7
ADW 053	68.2	73.5	80.5	9.5
DMA 066	66.2	70.9	101.0	1.5
DMA 002	64.5	69.7	90.5	-10.0
OFF 146	66.3	72.5	52.5	9.0
NN 42	65.9	70.4	127.5	8.0
ADW 063	65.6	71.3	123.0	22.0
OHYEOKA	66.8	70.8	98.0	15.5
ADEHYE	64.7	69.4	77.5	7.0
AFS 027	66.1	70.8	97.5	7.0
AFS 126	65.8	70.0	92.0	15.0
ADW 004	65.7	72.0	107.0	2.0
ADW 051	65.9	70.0	46.5	5.5
UCC 517	64.1	68.6	82.0	-11.5
AFS 041	65.0	71.0	67.5	-15.5
OFF 019	66.6	70.6	85.0	-5.5
OFF025	65.3	71.7	82.0	10.5
CAPEVARs*	65.9	69.9	94.0	-22.5
OFF 145	66.9	70.9	56.0	13.0
UCC 470	65.3	69.5	118.0	-7.0
UCC 506	65.8	71.2	21.5	-13.5

Table 18 continued

KW 161	66.2	72.6	83.5	6.5
AFS 136	67.3	72.1	123.0	14.5
ASAMAN	65.5	70.7	109.0	15.5
AFS 001	67.6	73.1	90.0	15.0
OFF 058	66.1	71.8	99.5	21.0
B.BOTAN*	67.0	71.6	140.0	17.5
NKABOM*	66.6	71.1	59.0	6.5
OFF 093	66.1	71.8	47.0	6.5
NN43	64.3	69.5	71.0	3.0
OFF063	65.2	70.2	121.0	5.0
OFF086	63.8	67.3	92.0	8.0
KW85	65.7	70.1	109.0	13.0
AFS131	65.1	69.5	77.0	-5.0
OFF136	64.8	70.0	94.0	-10.0
Mean	65.8	70.7	87.7	3.5
Range	63.8 - 68.2	67.3 - 73.5	21.5 - 140.0	-28.0 - 22.0
CV	1.4	1.7	29.2	348.6
Lsd	1.73	2.57	44.4	36.62

*Released varieties.

Viscosity characteristics

Peak viscosity

Table 19 shows the viscosity characteristics of different starches extracted from 43 cassava genotypes. The peak viscosity averaged at 643.7 BU, with a wide range of variation from 412.5 BU recorded by accession 'OFF 093' starch to 857.0 BU recorded by accession 'AFS 136' starch. Starches from 25 % of the cassava genotypes recorded peak viscosity less than 600 BU.

Breakdown viscosity

Starches from all the cassava genotype registered an average breakdown viscosity of 443.1 BU, with a range from 300.5 BU to 659.0 BU. Starch from accessions 'AFS 093' and 'ADW 004' recorded the lowest and highest values respectively.

Setback viscosity

Starch from accession 'AFS 136' registered the highest setback viscosity with the value of 200.5BU while accession 'OFF 093' gave the lowest value of 62.0 BU. The mean setback viscosity was 141.9 BU. Accessions 'DMA 002', 'AFS 027' and 'ADW 004' recorded setback viscosity of 142.0 BU.

Viscosity at 95 °C

Starch viscosity at 95 °C for the entire samples recorded a mean value of 288.2 BU, with a range of 159.0 to 426.5 BU. Starch from accessions 'AFS 136' and 'OFF 093' recorded the highest and the lowest values respectively. Starch from 18 cassava genotypes recorded viscosity at 95 °C values above the mean value.

Viscosity 15 minutes at 15 °C

The range of values recorded for viscosity at 95 °C after 15 minutes was from low 112.0 BU to a high value of 303.5 BU for 'OFF 093' and 'AFS 136' respectively. The average value recorded was 200.5 BU.

Ease of cooking (min)

The maximum duration required to cook starch from the various cassava genotypes was recorded by 'Asaman' (5.6 min), while the shortest duration was recorded by Nkabom (1.2 min). The average time to cook starch was recorded as 3.0 min.

Rankings in terms of peak viscosity, Setback viscosity and viscosity at 95 °C

Table 20 gives the ranks of 40 cassava accessions and 3 varieties in terms of peak viscosity, setback viscosity and viscosity at 95 °C. The overall 15 outstanding cassava genotypes in terms of peak viscosity, setback viscosity and viscosity at 95°C were 'OFF 029', 'DMA 002', 'AFS 027', 'OFF 058', 'ADW 063', 'AFS 136', 'ADW 004', 'KW85', 'AFS 048' and 'KW 161'. The rest were 'ASAMAN', 'DMA066', 'OFF023', 'AFS041' and Bankye Botan.

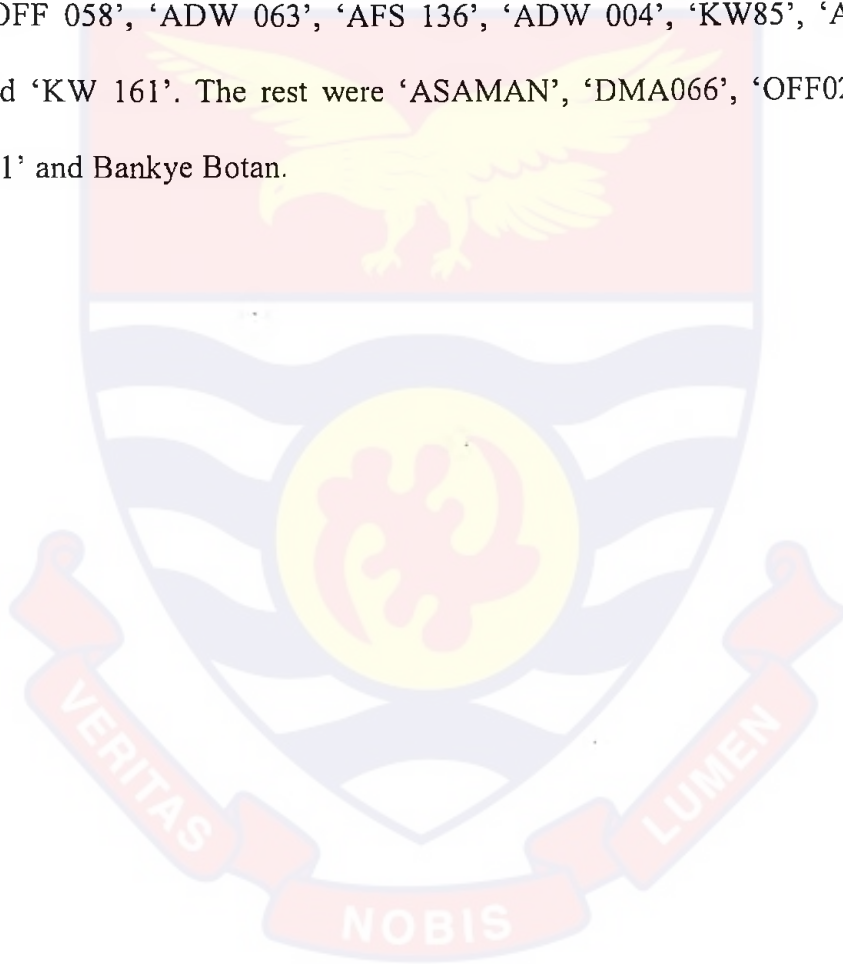


Table 19: Viscosity characteristics of starch from 40 cassava accessions and 3 varieties

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Accessions/ Varieties	Peak Viscosity (BU)	Breakdown Viscosity (BU)	Setback Viscosity (BU)	Viscosity at 95°C (BU)	Viscosity 15min at 95 °C (BU)	Ease of cooking (min)
BESEREBEMA	572.5	394.0	103.0	255.5	178.5	2.0
KW 148	733.5	508.5	176.0	299.5	225.0	4.4
KW 070	636.5	464.5	117.0	253.5	172.0	3.4
AFS 048	813.0	547.5	157.0	318.0	265.5	3.3
UCC 153	615.5	363.5	158.0	302.0	252.0	3.0
KW 001	600.5	417.0	186.0	265.5	183.5	2.8
KW 181	626.0	435.5	146.5	306.0	190.5	3.5
OFF 029	735.5	527.5	129.0	312.5	208.0	2.4
OFF 023	734.0	551.0	154.0	300.0	183.0	3.1
ADW 053	581.0	376.5	128.5	285.0	204.5	3.0
DMA 066	705.0	471.0	159.0	335.0	234.0	3.0
DMA 002	748.0	506.5	141.0	332.0	241.5	4.7
OFF 146	477.5	334.5	81.5	195.5	143.0	4.7
NN 42	618.5	452.5	147.5	293.5	166.0	3.0
ADW 063	657.5	459.5	135.5	321.0	198.0	4.0
OHYEOKA	584.0	370.0	128.5	312.0	214.0	3.1
ADEHYE	534.0	343.5	125.0	268.0	190.5	2.7
AFS 027	751.5	525.5	141.0	323.5	226.0	4.3
AFS 126	644.0	449.0	162.0	287.0	195.0	2.4
ADW 004	849.0	659.0	141.0	337.0	190.0	3.7
ADW 051	639.0	471.0	159.5	214.5	168.0	2.6
UCC 517	627.5	453.0	117.0	256.5	174.5	1.6

Table 19 continued

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AFS 041	647.5	408.0	140.0	307.0	239.5	3.2
OFF 019	622.5	451.5	155.0	256.0	171.0	2.3
OFF025	622.5	448.5	166.5	256.0	174.0	2.1
CAPEVARs*	506.5	353.5	137.5	247.0	153.0	2.7
OFF 145	681.5	479.0	135.0	258.5	202.5	2.8
UCC 470	581.5	402.0	148.5	297.5	179.5	2.8
UCC 506	648.0	467.0	104.5	202.5	181.0	2.6
KW 161	708.0	471.0	149.0	320.5	237.0	3.9
AFS 136	857.0	553.5	200.5	426.5	303.5	4.0
ASAMAN	550.0	324.0	117.5	297.0	226.0	5.6
AFS 001	669.5	412.0	163.0	347.5	257.5	4.7
OFF 058	726.0	499.0	137.0	326.5	227.0	4.0
B. BOTAN*	721.5	463.0	189.0	398.5	258.5	4.5
NKABOM*	547.5	390.0	157.0	216.5	157.5	1.2
OFF 093	412.5	300.5	62.0	159.0	112.0	2.2
NN43	621.0	501.0	111.0	191.0	120.0	3.0
OFF063	589.0	389.0	134.0	321.0	200.0	1.5
OFF086	615.0	454.0	156.0	253.0	161.0	3.1
KW85	687.0	502.0	124.0	294.0	185.0	2.6
AFS131	608.0	397.0	163.0	288.0	211.0	1.8
OFF136	573.0	309.0	157.0	358.0	264.0	3.5
Mean	643.7	443.1	141.9	288.2	200.5	3.0
Range	412.5 - 857.0	300.5 - 659.0	62.0 - 200.5	159.0 - 426.5	112.0 - 303.5	1.2 - 5.6
%CV	14.2	16.8	18.8	18.4	20.0	33.3
Lsd	95.94	85.31	59.69	63.66	48.95	0.49

*Released varieties

Table 20: Rankings of 40 cassava accessions and 3 varieties in terms of peak viscosity, Setback viscosity and viscosity at 95 °C

Accession/varieties	Peak viscosity	Setback viscosity	Viscosity at 95°C	Rank sum	Overall
OFF 029	6	13	14	33	1
DMA 002	5	21	7	33	2
AFS 027	4	21	9	34	3
OFF 058	9	17	8	34	4
ADW 063	16	16	11	43	5
AFS 136	1	43	1	45	6
ADW 004	2	21	22	45	7
KW85	13	9	23	45	8
AFS 048	3	31	13	47	9
KW 161	11	26	12	49	10
ASAMAN	38	8	5	51	11
DMA 066	12	34	6	52	12
OFF 023	7	27	19	53	13
AFS 041	18	19	16	53	14
BANKYE BOTAN*	10	42	2	54	15
OFF063	32	14	10	56	16
AFS 001	15	37.5	4	56.5	17
OFF 145	14	15	30	59	18
UCC 517	22	6.5	31	59.5	19
OHYEOKA	33	11.5	15	59.5	20
UCC 506	17	4	40	61	21
KW 070	21	6.5	35	62.5	22
KW 181	23	23	17	63	23
KW 148	8	40	20	68	24
OFF136	36	31	3	70	25
NN43	26	5	42	73	26
ADW 053	35	11.5	27	73.5	27
BESEREBEMA	37	3	34	74	28
NN 42	27	24	24	75	29
ADEHYE	40	10	28	78	30
UCC 153	28	33	18	79	31
UCC 470	34	25	21	80	32
AFS 126	19	36	26	81	33

Table 20 continued

OFF 019	24	28	32	84	34
OFF 146	42	2	41	85	35
OFF 093	43	1	43	87	36
ADW 051	20	35	39	94	38
OFF086	29	29	36	94	39
CAPEVARs*	41	18	37	96	40
OFF025	25	39	33	97	41
KW 001	31	41	29	101	42
NKABOM*	39	31	38	108	43

*Released varieties

Functional properties of starch mixtures

Swelling volume

Swelling volume of starch mixtures of Adehye and sweet potato (Ad:Sp) and that of AFS 048 and sweet potato (AFS:Sp) decrease linearly as the proportion of sweet potato starch increases. However, OFF 146 and sweet potato (OFF:Sp) and Bankye Botan and sweet potato (BB:Sp) did not show any clear pattern (Table 21).

Swelling power (g g^{-1})

Starch mixtures ((Ad:Sp) and (AFS:Sp)) exhibited linear change in values as the starches were mixed together. Both mixtures recorded linear decrease in swelling power as percentage of sweet potato starch in the sample increases. However, starch mixtures ((OFF:Sp) and (BB:Sp)) did not show any clear pattern. Besides, the value for these mixtures were higher than that of sweet potato values recorded.

Solubility (%)

The values recorded for all the starch mixtures did not show any regular pattern.

Pasting and viscosity characteristics of starch mixtures

The pasting and viscosity characteristics of cassava and sweet potato starch mixtures are presented in Table 22. Pasting and peak temperature exhibited a linear increase pattern. The pasting and peak temperatures of starch from cassava increased as the proportion of sweet potato in the mixture increases.

Table 21: Functional Properties of Cassava Starch and Sweet Potato Starch mixtures

Sample	Swelling Volume (mL g ⁻¹)	Swelling Power (g g ⁻¹)	Solubility (%)
Cassava(Adehye)	32.2	35.1	10.3
C ₈ Sp ₂	24.5	30.7	15.0
C ₅ Sp ₅	23.3	27.1	16.0
C ₂ Sp ₈	22.2	25.9	14.0
Sweet potato	20.7	23.3	12.3
Cassava(OFF146)	23.0	30.5	27.7
C ₈ Sp ₂	26.8	31.3	13.0
C ₅ Sp ₅	25.2	29.1	11.3
C ₂ Sp ₈	21.7	24.7	12.7
Sweet potato	20.7	23.3	12.3
Cassava (B. Botan)	29.3	35.3	16.3
C ₈ Sp ₂	24.5	30.7	15.0
C ₅ Sp ₅	24.8	31.5	22.7
C ₂ Sp ₈	22.3	26.4	16.3
Sweet potato	20.7	23.3	12.3

Table 21 continued

Cassava (AFS 048)	27.3	28.5	11.7
C ₈ Sp ₂	26.2	31.2	15.4
C ₅ Sp ₅	25.7	28.4	14.0
C ₂ Sp ₈	22.3	24.8	11.3
Sweet potato	20.7	23.3	12.3

C₈SP₂ = 80% cassava: 20% sweet potato

C₅SP₅ = 50% cassava: 50% sweet potato

C₂SP₈ = 20% cassava: 80% sweet potato

Ease of cooking was appreciably shorter for OFF:Sp mixtures, while BB:Sp mixtures recorded the highest values. Ease of cooking of cassava starch was prolonged/enhanced by the addition of sweet potato starch. It increased as the proportion of sweet potato was increased recording the highest values at 50:50 ratios. However, beyond the 50:50 ratios the duration reduced or shortened.

Peak viscosity, breakdown, set back, paste stability at 95 °C and paste stability at 50 °C did not show any clear pattern. However, with the exception of C₈Sp₂ (BB:Sp) mixture, the remaining C₈Sp₂ mixtures experienced an increase in paste stability at 50 °C.

Table 22: Pasting and viscosity characteristic of cassava and sweet potato starch mixtures

Samples	Pasting Temp. (°C)	Peak Temp. (°C)	© University of Cape Coast https://ir.ucc.edu.gh/xmlui Ease of cooking (Min)	Peak Viscosity (BU)	Breakdown Viscosity (BU)	Setback Viscosity (BU)	Paste Stability at 95°C (BU)	Paste Stability at 50°C (BU)
Cassava(Adehye)	64.7	69.4	3.4	534.0	343.5	125.0	77.5	7.0
C ₈ SP ₂	64.5	76.7	6.7	707.5	426.5	206.0	123.0	12.0
C ₅ SP ₅	65.9	79.0	8.6	571.0	322.5	168.5	138.5	7.5
C ₂ SP ₈	75.8	81.6	3.3	600.5	222.5	128.5	78.0	14.5
Sweet Potato	76.8	82.0	2.5	645.5	284.0	148.5	95.0	15.5
Cassava(OFF146)	66.3	72.5	4.7	477.5	334.5	81.5	52.5	9.0
C ₈ SP ₂	70.8	79.0	5.2	679.0	33.5	193.5	75.5	22.0
C ₅ SP ₅	67.9	78.6	6.1	580.0	310.5	146.5	62.0	16.5
C ₂ SP ₈	75.3	80.1	3.0	607.0	270.5	143.5	82.5	20.0
Sweet Potato	76.8	82.0	2.5	645.5	284.0	148.5	95.0	15.5
Cassava(B.Botan)	67.0	71.6	4.5	721.5	463.0	189.0	140.0	17.5
C ₈ SP ₂	65.4	76.9	8.0	540.5	334.0	123.5	93.0	-7.0
C ₅ SP ₅	66.0	79.1	9.2	557.0	325.0	148.0	94.0	2.5
C ₂ SP ₈	76.3	81.4	3.4	604.5	230.5	128.0	91.0	5.5
Sweet Potato	76.8	82.0	2.5	645.5	284.0	148.5	95.0	15.5
Cassava(AFS048)	64.6	70.3	3.3	813.0	547.5	157.0	52.5	-28.0
C ₈ SP ₂	64.2	77.0	7.5	675.0	436.5	173.0	129.0	4.5
C ₅ SP ₅	65.0	78.4	7.9	615.0	323.5	145.5	94.5	4.5
C ₂ SP ₈	75.9	79.7	1.8	630.5	246.5	166.0	87.5	11.0
Sweet Potato	76.8	82.0	2.5	645.5	284.0	148.5	95.0	15.5

C₈SP₂ = 80% cassava: 20% sweet potato

C₅SP₅ = 50% cassava: 50% sweet potato

C₂SP₈ = 20% cassava: 80% sweet potato

Table 23 gives the ranks of 40 cassava accessions and 3 varieties in terms of mosaic severity score, % harvest index, tuber yield and starch yield characteristics. The 10 genotypes that were least affected by ACMV disease were: 'Cape Vars', 'Adehye', 'KW 085', 'OFF 063', 'B. BOTAN', 'UCC 470', 'NKABOM', 'AFS 041', 'AFS 048' and 'OFF 136'. The top ten ranking genotypes that recorded high percentage harvest index were: 'Adehye', 'ADW 051', 'ADW 053', 'B. BOTAN', 'UCC 517', 'AFS 027', 'NN 43', 'OFF 145', 'OFF 023' and 'AFS 131'. 'KW148', 'KW 001', 'UCC 153', 'OFF 063', 'AFS 041', 'ADW 053', 'ADW 051', 'OFF 136', 'AFS 001' and 'ADEHYE' were the superior tuber yielding genotypes. The ten outstanding starch yielders were: 'AFS 048', 'UCC 470', 'ADW 053', 'KW 001', 'KW 148', 'OFF 019', 'OFF 145', 'UCC 517', 'AFS 136' and 'B. BOTAN'.

Table 23: Rankings of 40 cassava accessions and 3 varieties in terms of mosaic severity score, % harvest index, tuber yield and starch yield characteristics

Accessions/ varieties	Mosaic Severity score	Harvest Index (%)	Tuber yield (t ha ⁻¹)	Starch content (%)	Rank sum	Overall
ADEHYE	2	1	10	15	28	1
ADW 053	18	3	6	3	30	2.5
B. BOTAN*	5	4	11	10	30	2.5
KW 148	16	19	1	5	41	4
KW 001	14	22	2	4	42	5
NKABOM*	7	15	15	12	49	6.5
OFF 019	13	12	18	6	49	6.5
ADW 051	17	2	7	26	52	8
CAPEVARs*	1	11	21	22	55	9
AFS 041	8	32	5	11	56	10.5
UCC 517	19	5	24	8	56	10.5
UCC 470	6	27	22	2	57	12
OFF 145	29	8	16	7	60	13
OFF 063	4	17	4	36	61	14.5

Table 23 continued

UCC 153	11	14	3	33	61	14.5
AFS 136	20	21	12	9	62	16
OFF 136	10	31	8	17	66	17
AFS 048	9	36	26	1	72	18.5
OFF 023	15	9	19	29	72	18.5
UCC506	12	13	13	35	73	20
AFS 131	36	10	14	14	74	21
AFS 027	28	6	31	13	78	22.5
AFS 001	33	18	9	18	78	22.5
KW 161	22	20	17	27	86	24
DMA 066	26	24	25	20	95	25.5
OFF 058	27	29	23	16	95	25.5
BESEREBEMA	25	25	32	19	101	27
NN43	37	7	20	43	107	28
OHYEOKA	32	16	29	31	108	29
KW 085	3	43	41	25	112	30
DMA 002	41	23	27	24	115	31
ASAMAN	38	30	28	23	119	32
NN 42	23	28	34	39	124	33
KW 181	31	26	33	38	128	34
OFF 093	24	40	40	28	132	35
ADW 004	42	35	38	21	136	36
OFF 146	34	33	30	41	138	37
OFF 086	21	39	39	40	139	38
ADW 063	30	34	35	42	141	39
AFS 126	40	37	37	30	144	40
OFF 025	35	38	36	37	146	41
KW 070	39	41	43	32	155	42
OFF 029	43	42	42	34	161	43

*Released varieties

Rank: 1= highest... 43 = lowest

Overall rank: lowest = best; highest = worst

The overall outstanding cassava genotypes in terms of resistance to mosaic, percentage harvest index, tuber yield, and starch yield were: 'ADEHYE', 'ADW 053', 'B. BOTAN', 'KW 148', 'KW 001', 'NKABOM', 'OFF 019', 'ADW 051', 'CAPE VARS' and 'AFS 041'.

CHAPTER FIVE

DISCUSSION

Variation in qualitative characters

Qualitative characters are very important in the identification of crops due to their relative stability over the quantitative characters. Variation in the leaf characters was studied using the first leaf, leaf vein, petiole colour and pubescence of young leaf. Within the total accessions light green emerged as the most predominant character among the first leaf. It was followed by dark green, with purple being the character with the lowest relative frequency. Cultivars with purple petiole colours predominated in the total accession. Doku (1969) observed the following cassava petiole colours: uniform purple, red, pink, maroon or various shades of these on both surfaces, and upper surface coloured with freckled red or purple. Ampong-Mensah (2000) after studying 400 accessions of cassava from Western and Central Regions of Ghana observed light green, green purple and purple as petiole colours. In this present studies, light green, green purple and purple petiole colours were observed in agreement with Ampong-Mensah (2000), while red, pink and maroon types were absent as reported by Doku (1969). On the other hand purple emerged as the dominant character among the leaf vein colours in the total accessions.

Petiole length could be a measure of the width of canopy formed by a cassava plant and the extent to which the associated crop in an intercrop

can be shaded. The prevalence of cultivars with long petiole length in this study suggests that most of the cultivars may not be suitable for intercropping, as they would reduce the reception of solar energy for the companion crops. According to El-Sharkawy et al. (1992), cassava is a crop that requires high solar radiation to perform photosynthesis more efficiently. This means that most of the accessions in this study could compete with associated crops such as maize which are usually planted earlier than cassava.

Cultivars with no young leaf pubescence recorded the highest relative frequency. This study agrees with the findings of Ampong-Mensah (2000). Pubescence of young leaves has been reported to be positively related to reduced cassava green mite (GM) and cassava mealy bug (CMB) (Hahn & Keyser, 1985; Kanno et al., 1991). Thus breeders who wish to upgrade their cultivars could use those that possess this trait as parental materials.

Stem colour is one of the three most stable vegetative characters (petiole, stem and cortex) normally used in classifying cassava (Doku, 1969, Oduro et al., 1989; Ampong-Mensah 2000). Light brown cultivars predominated, followed by dark brown and silver green. The traits, which seem to be selected for might probably, be related to some of the desirable traits (early maturity, poundability, etc) preferred by farmers.

Root characters are known to be the main determinant of a variety that would be selected by a farmer. The reason had been that roots form the economic important part of the cassava plant in Ghana. Three classes of root surface colour were observed. The brown coloured (light or dark) root

surface cultivars seemed to have been selected for by the farmers/cultivators in cassava growing areas as against the white/cream ones. The result of this study revealed dark brown as the predominant colour and was consistent with report made by Onwueme (1978) and Ampong-Mensah (2000) that brown was the predominant tuber surface colour.

Root cortex outer surface colour is also one of the most important traits used to describe cassava varieties (Oduro et al., 1989). In Ghana the trait plays a key role in the selection criteria by cultivators and consumers alike, as they perceive that this trait is highly associated with mealiness of cassava varieties (Ampong-Mensah, 2000). In this study, white/cream cortex surface predominated as the root cortex outer surface colour, followed by purple while pink and yellow had the lowest frequencies. It thus appeared that cultivars with white/cream and purple root cortex outer surface colours were capable of providing the desired food characteristics and agronomic needs of the cultivators and consumers and hence has been selected for. Pink and yellow coloured root cortex cultivars on the other hand, might either be under selection pressure.

The existence of two types of cassava root pulp colour, white/cream and yellow in Africa has been reported (Dade, 1930; Doku 1969; Oduro et al., 1989; Ampong-Mensah, 2000). The result of this study indicated that the cassava germplasm collections were largely composed of white/cream root fresh types. Even though, cultivars with yellow coloured root pulp are known to have high levels of β -carotene, a vitamin A precursor (McDowell & Oduro 1983; Maravalhas, 1984). There is the need to further evaluate

the cultivars with this trait to ascertain reasons for the low frequency recorded in this study. The result suggests that the yellow coloured pulp cultivars are being genetically eroded.

Variation in quantitative characters

Quantitative traits are controlled by several factors and hence are not stable as qualitative traits. For any crop improvement program aimed at achieving maximum yield, knowledge of genetic variability of several traits and their contribution to yield is essential. Thus knowledge of parameters such as range, mean and coefficient of variation are basic in the genetic improvement of a crop.

The coefficient of variation (CV) showed large difference among the characters studied. The CV ranged between 4.7% and 158.8%. Petiole length, plant height central leaf lobe length, central leaf lobe width and storage root length recorded low CV of 4.7 %, 19.0 %, 23.2 %, 23.2 % and 23.3 % respectively. This indicates that these traits showed limited variability among which petiole length showed the least. Birador et al. (1978) and Ampong-Mensah, (2000) also recorded low genetic coefficient of variation for plant height. Central leaf lobe length and width recorded the same CV of 23.2 %. The less variation among the accession for the characters suggests that there is less potential for improving these traits, especially petiole colour, as breeders would have less variation to operate on.

Number of level of true branching, first apical branch height, level of lateral branching, height at first lateral branching and number of rotten

storage root/plant on the other hand, showed high CV. Among these traits, the highest CVs were observed for number of rotten storage root/plant (158.8 %), followed by level of lateral branching (85.5 %) and level of true branching (64.0 %). These suggest that there were wide variations among the accessions with respect to branching habit (true and lateral branching) indicating a better scope of selection for these characters in breeding programs. However, an accession with profuse lateral branching is undesirable for selection because it makes cultural practice operations difficult to carry out, makes the stems unattractive for planting and also hinders mechanized farming.

Morphological characterization

Morphological traits are useful for preliminary evaluation, because they offer a fast and easy approach for assessing the extent of diversity. The estimation of descriptive statistics of 19 different morphological traits studied in the present study revealed the existence of high level of morphological diversity among the cassava accessions, providing room for improvement through hybridization and selection. Carvalho and Schaal (2001) identified a high degree of genetic variability among 94 cassava accessions of Brazilian origin. Factor analysis was performed in order to reduce a set of morphological traits (19) to a more meaningful smaller set of traits (14) and to identify the trait contributing for maximum variability, since it provides an exact picture of variability contributed by each trait. On the basis of factor loading of the 19 morphological traits, first 3 factors contributing to variability are selected for principal component analysis.

While using the traits (which accounted for maximum variation in the factor analysis) in the principal component analysis, central lobe length, petiole colour, colour of root cortex, yield per plant, petiole length, and root length showed the maximum variability in the total variation. The genetic diversity analysed using morphological traits showed that there was a high amount of variation for the quantitative traits.

In cluster analysis the similarity matrix was computed using morphological markers based on Nei's coefficient following the UPGMA method using PowerMarker version 3.25. Nei's similarity coefficient for morphological data and SSR data varied from 0.00 to 0.40 and 0.00 to 0.25 respectively. Based on morphological data, the 43 cassava accessions were basically grouped into two broad clusters however, at 0.15 similarity coefficient they were sub clustered into 4. Among these different clusters, the cluster size varied from 5 for cluster I to 18 for cluster IV (Figure 12). The II and III clusters had 10 members each. Adehye and Capevars were closely related in cluster II. All the released varieties, Bankye Botan, Capevars and Nkabom, were found in different clusters - I, III and IV respectively, thus indicating that they were morphologically different from each other.

Stem colour, colour of young leaf, colour of root cortex, leaf vein colour, petiole length, yield and root surface colour were more variable among the accessions than the other traits (pubescence of young leaves, plant height, petiole length, pulp colour and root diameter).

Both qualitative and quantitative morphological traits are valuable tools for cassava characterization. Many researchers have carried out

studies on cassava using morphological traits like stem girth, tuber length, tuber girth, yield per plant, tuber attachment on parent cuttings, tuber inner skin colour, tuber outer skin colour, tuber surface texture and pulp colour and yield (Ampong-Mensah 2000; Fregene et al., 2000; Carvalho & Schaal, 2001; Elias et al., 2001; Kizito et al., 2006; Raghu et al., 2007). These traits were all found to be of great importance in distinguishing germplasm leading to better classification. However, morphological traits are known to be influenced by external environment resulting in varying relationship pattern (Smith & Smith, 1989), hence the argument for DNA molecular markers in genetic diversity studies which remain constant with changes in the environment.

Molecular characterization

Molecular diversity was assessed by using 20 SSR primers. Number of allele produced by different primers ranged from 2 to 9 with an average of 4.9 alleles per primer. Among the primers used, SSRY20, and SSRY175 produced a maximum of 9 alleles, while SSRY103 and SSRY21 also followed with 8 alleles. Similarly, Raghu et al. (2007) studying Indian cassava accessions with 15 SSR primers recorded mean number of 4 alleles with a range of 2 to 6 alleles. Microsatellite markers generated a higher number of allele while analyzing the Central American cassava accessions (Chavarriaga-Aguirre et al., 1999).

SSR primers in general have shown high levels of polymorphism in many important crops including *Oryza sativa* L. (Bligh et al., 1999), *Sorghum bicolor* (Smith et al., 2000), *Vigna unguiculata* L (Chen-Dao

et al., 2001), *Vitis vinifera*, (Di Gaspero et al., 2000), *Prunus pandorea* and *Prunus amygdalus* (Ajay et al., 1999), *Triticum aestivum* L (Ahmad, 2002), *Helianthus annuus* L., (Yu et al., 2000), *Cucumis melon* L (Danin-Poleg et al., 2001) and *Manihot esculanta* (Mba et al., 2000; Moyib et al., 2007; Raghu et al., 2007). The mean PIC value recorded in the current study compared favourably with results obtained from another studies by Tams et al. (2004), where a study of 128 accessions of Triticale with 28 SSR markers gave a mean PIC value of 0.54. The result of this study therefore showed that all the primers were highly informative and can be used for genetic diversity studies and the study of phylogenetic relationship.

The computed heterozygosity values were high. The high observed heterozygosity values in this study corroborate the heterozygote nature of most of the accessions and the fact that cassava is largely cross-pollinated. The negative values of the inbreeding coefficient ($f = -0.31$) as shown in Table 10 further confirms the heterozygosity among accessions.

SSR marker profiles resulted also resulted in two broad clusters. However, 9 sub clusters were obtained at 0.15 similarity coefficient. The similarity matrix coefficient of the molecular data ranged from 0.00 to 0.25 and that of the morphological data ranged from 0.00 to 0.40. This indicated that the genetic diversity observed using molecular data is higher than that observed using morphological data. This resulted in sub clusters of 4 and 9 at similarity coefficient of 0.15 for morphological data and SSR marker profile respectively. Raghu et al, (2007) recorded similar findings when they studied 58 cassava accessions with 29 morphological traits and 15 SSR primers. They recorded similarity coefficient of 0.29 to 0.59 for SSR

data and 0.48 to 1 for morphological data; with morphological data grouping the 58 accessions into 6 and molecular data grouping them into 9 clusters. This study confirms the fact that SSR markers are able to distinguish accessions properly than morphological markers. Genes are stable while morphological traits are affected by the environment (Smith & Smith, 1989; Gepts, 1993; Raghu et al., 2007).

Generally, the relationship between accessions within the clusters could not be attributed to their origin, since accessions from various locations clustered together. The Adehye and Capevars accessions were in the same clusters for both morphological and molecular traits, thus reflecting their genetic similarity. The inability of the markers to distinguish between Adehye and Capevars accessions, AFS048 and OFF063 as well as Asaman and Beserebema indicates that these are duplicates in the collection. Lokko et al. (2005) identified TME240 and TME242 from Togo and TME4 and TME62 as duplicates in their collection. Detection of duplicates in a collection is critical for effective management of germplasm. In general, many of the genotypes were clustered together, with very few, such as Ohyeoka, AFS027 and OFF058 in distinct clusters based on SSR data.

Whiteflies, mosaic severity and plant yield

The whitefly, *Bemisia tabaci*, is one of the most important insect pests in world agriculture because of its direct feeding, contamination from honeydew, and ability to transmit plant viruses (Perrings, 2001). Additional evidence of differences in whitefly infestation among a range of cassava

genotypes at different locations in Ghana were also found in the present study.

The adult whitefly population was high in the 6th week after planting (WAP) in both years. Generally the whitefly population increased in 2008 compared to 2007 and this might be due to increased rainfall pattern and the marginal decline in temperature (Table 4), this agrees with the findings of Legg et al. (2003). The highest mean severity score for 2007 was recorded at 12 WAP. This finding agrees with Leuschner (1978) and Ogbe et al. (1996) that high incidence of CMD is achieved at 12 WAP. However, in 2008 the highest mean severity was recorded at 6 WAP. It might be due to the fact that the same accessions were used which might have been infested already. This confirms the reports of Fargette et al. (1988) that plants are generally more susceptible to secondary infection.

A higher number of whiteflies were found on resistant accessions in this study, which agrees with Otim et al. (2005) who recorded higher populations of *B. tabaci* on the cassava mosaic disease-resistant accessions compared to susceptible ones. Similar observations have been made by Legg et al. (2003), and are attributed to the whitefly preference for the resistant varieties of cassava. The leaves of resistant plants were broader and softer than the susceptible ones, whose leaves were misshapen, highly reduced and showed severe mosaic symptoms. According to Sserubombwe et al. (2001), Omongo (2003) and Ariyo et al. (2005), such leaves are usually avoided by the whitefly and this might account for the whitefly preference for the resistant plants in this study.

Otim-Nape et al. (1994) has also reported the lack of any significant correlation between whitefly numbers and mosaic severity when they studied the effects of African cassava mosaic geminivirus on the main cassava varieties grown in three districts of western Uganda. They argued that the lack of significant correlation between the two variables was probably due to infection of the plants with symptoms sometime before the whitefly counts were made and that many of the plants were likely to have been established from infected cuttings. Similar explanation could be assigned to the observed lack of significant correlation between whitefly populations and mosaic severity in the current study, since the cassava genotypes tested were obtained from diverse origins and, therefore, could not be considered entirely free of ACMV disease infection.

Although mixed infections of ACMV and EACMV have been reported in West Africa (Ogbe et al., 1999; Offei et al., 1999), the results of this study showed that the only CMG strain responsible for the disease symptom was ACMV. The ACMV primer ACMV-1/ACMV-2 was more efficient in detecting the virus in the samples in that it detected the virus in more samples than the rest of mosaic primers.

The presence of the virus in some of the resistant samples suggests that field resistance observed as no symptoms, was not necessarily an indication of resistance to virus infection. The A genome of geminiviruses of which the cassava mosaic viruses belong, encode a protein required for their replication and must recruit the remaining DNA replication mechanism from the host plant, while the B genome is responsible for spread and symptom production (Estessami et al., 1991; Fontes et al.,

1992). Since DNA replication is part of the natural growth and development, it is possible that the virus is able to replicate and probably even spread in the resistant plant but the subsequent disease symptoms are inhibited. Ogbe (2001) has concluded that field resistance, as shown by lack of symptoms, is not necessarily an indication of resistance to virus infection, but may be partly due to lack of virus multiplication, which suggests that field selection of resistance should be complemented with PCR or inoculation test.

The presences of band for scar marker for CMD2 gene in the four selected genotypes suggest that CMD2 gene was responsible for the resistance in the four genotypes. However the presence of bands in Capevar, Adehye, and KW058 with Nkabom which showed very mild mosaic symptoms also suggest that the scar markers used were not close to CMD2 gene in those accessions and that other source of resistance should be exploited. It is also possible that some strains have evolved as satellite virus which may require a helper virus to express its symptoms and this may account for presence of bands in some of the resistant accessions.

Yield and yield components

The number of roots per plant varied between 0.3 and 9.4 and the range of these figures is higher than the values recorded by Adomako (2007). He recorded values between 1.2 and 2.89 when he studied 11 cassava accessions at two locations. The values recorded in this study however compare favourably to what Otim-Nape et al. (1994) have reported after studying 13 cassava varieties in three districts of western

Uganda. They reported a range of 1.0 to 10.0 roots per plant. The differences in the number of roots per plant which were significant ($P < 0.05$) in this work could be attributed to varietal differences. Pursglove, (1987) and IITA, (1990) reported that 4-10 tubers per plant may be produced.

Magoon et al. (1970) after studying a large number of F_1 cassava plants obtained from crosses among three plants of diverse origin found out that yield was closely related to the number of storage roots (range 1 to 12). It is therefore likely that cassava genotypes identified in this work with relatively large root numbers per plant may produce higher yields. Accessions 'UCC517', 'AFS041', 'ADW053', 'UCC506' and 'KW148' recorded the highest number of roots per plant and therefore they are likely to give higher root yields.

Fresh root weight recorded in this study ranged from 0.1 to 9.4 kg. Fresh root weights recorded in this experiment were relatively higher in values than what had been reported in literature. For instance, Fargette et al. (1988) in Côte d'Ivoire and Terry and Hahn (1990) in Nigeria have found that when harvested after 12 months cassava yields per plant were 2.4 – 5.2 kg and 1.4 – 3.0 kg respectively. The differences in the fresh root weights per plant were significant ($p < 0.05$). Fresh weight recorded CV of 66.1 %. This indicates that there was some extent of variation among the accessions as far as this trait was concerned. This is useful information to the breeders since it indicates diversity among the crop from which selection could be made for further improvement.

The highest fresh root yield recorded in this experiment was 93.8 t ha⁻¹ and the lowest was 1.0 t ha⁻¹ produced by accession 'KW148' and accession 'KW070' respectively. The mean value of 31.02t/ha was recorded. The values recorded in this study are higher than values recorded elsewhere. Chitiyo and Kasele (2004) studied 18 new introduced cassava varieties and two local accessions at two locations in Zimbabwe and reported a fresh root yield range of 2.7 to 12.9 t ha⁻¹. Nweke et al. (1994) surveyed 501 cassava farms in Southeast Nigeria and estimated that the overall mean of fresh root yield was 11.9 t/ha. These reported yields indicate that the yields obtained in this work are relatively high. However, this result compared fairly with work done by Afuakwa et al. (1999) on four cassava genotypes, namely: 'Gblemoduade', 'Abasafitaa', 'Tekbankye' and 'Afisiasi' showed the average fresh root yield at 12 months maturity to be 33-38 t ha⁻¹; 26-31 t ha⁻¹; 26-31 t/ha and 27-30 t ha⁻¹ respectively. When the yield components, that is, number of roots per plant, fresh root weight (kg), individual root weight (g) and harvest index were examined it was observed that all the variables indicated significant (P<0.05) treatment mean differences. This result confirms the findings of Adomako (2007) except that he recorded no significant difference between the weights of individual tubers.

As has been explained by Cock et al. (1979) and Hunt (1990), harvest index is the ratio (quotient) between the weight of tubers or the marketable component of the crop to the weight of above-ground parts plus weight of tubers (total weight of crop) and hence cassava genotypes with less fresh shoot weights will show high harvest index values. Therefore the

high harvest index values registered by accessions 'Adehye' and 'ADW053' are accounted for by the less shoot weights recorded by these accessions and high root weight.

The harvest index recorded in this study ranged from 3.09 to 58.60 with the mean of 36.43. Harvest index values reported in this study are lower compared to result from other cassava genotypes elsewhere. For instance, Otim-Nape et al. (1994) produced harvest index data for thirteen cassava genotypes and ten out of this number showed a harvest index range of 40 to 80 and a mean of 60 and Nweke et al. (1994) stated a mean harvest index of 50 from 497 cassava farms surveyed.

The significant ($P < 0.05$) difference observed amongst the treatment effects may be attributed to genetic differences amongst the cassava genotypes as has been indicated by the studies of Pérez et al. (2001). As has been indicated earlier root yield is the economic part of cassava plant and important factor in determining the harvest index of the crop. Accession with higher harvest index shows that more assimilate is converted into root development than shoot. Harvest index is therefore significant in the selection of elite cassava genotypes in breeding programmes.

Most of the accession used in this study recorded low yield. According to Cock et al. (1979) few tubers per plant, low individual tuber weight and low harvest index which indicate poor partitioning or accumulation of assimilates in storage organs are attributes of a cassava plant that may result in low tuber yield. This may explain why relatively low fresh tuber weights were obtained for most of the accessions in this experiment.

Starch content and yield

The range of variation in starch content (%) values was between 14.64 and 24.83 %. These values fall within the range stated in literature elsewhere. Radley (1976) has stated that on average the tubers of cassava contain 20 – 30 % starch, but variations have been obtained as low as 12 % and as high as 33 % starch. Janssens (2001) has reported 20-40 % as the range of starch content in cassava tubers. In another work Adomako (2007) reported values between 23.07 and 25.01 %. No significant ($P>0.05$) differences were observed amongst the cassava genotypes tested with respect to starch content, which in essence, statistically means that the differences noted cannot be attributed to varietal differences.

Starch yield values were obtained by multiplying starch content (%) by fresh root weight ($t\ ha^{-1}$). Since starch content values were not significantly ($P>0.05$) different from each other, the significant ($P<0.05$) differences observed in starch yield were due to significant ($P<0.05$) differences detected amongst fresh root weights.

If starch yield is considered as an important factor in the selection of promising cassava genotypes, then 'KW148', 'KW001', and 'ADW 053', 'AFS 041', 'UCC 153', 'BANKYE BOTAN' and 'ADEHYE' stand out as the genotypes that should be selected.

Functional properties of starch

Swelling power provides evidence of non-covalent bonding between starch molecules; it is affected by factors such as amylose-

amylopectin ratio and chain length. Molecular weight distribution, degree of branching and conformation determine the degree of swelling and solubility (Rickard et al., 1991). The swelling volume values of the 43 cassava genotypes obtained in this work ranged between 14.5 and 35.3 mL g⁻¹. Adomako (2007) reported 16.30 and 26.17 mL g⁻¹ at Legon and between 17.0 and 28.67 mL g⁻¹ at Bunso. In another work, Moorthy (1994) studied the swelling behaviour of starch from eight cassava varieties and found out that the swelling volume of the different starches varied from 25.5 to 41.8 mL g⁻¹. The values obtained by Moorthy (1994) are relatively higher than what are reported here while the values obtained in this study on the other hand, are higher than what was reported by Adomako (2007). The significant ($P < 0.05$) differences in swelling volume values obtained in the work are indicative that swelling volume of cassava starch is dependent on variety among other factors as reported by Moorthy and Ramarujan (1986).

Solubility of starch depends on a number of factors such as inter-associative forces swelling power and the presence of other components. Cassava starch has a higher solubility than other tuber crop starches and the higher solubility can be attributed partly to the high swelling cassava starch undergoes during gelatinization (Moorthy, 2001). Percentage solubility recorded in this experiment varied from 8.3 to 27.5 %. Moorthy (1994) reported starch solubility values that ranged between 17.7 and 27.6 % when he studied the swelling behaviour of starch from 8 Indian cassava genotypes. The results obtained in this study compared favourably with Moorthy (1994).

Significant ($P < 0.05$) differences amongst the treatments were observed. These differences are attributable to varietal differences as has been postulated by Moorthy and Ramarujan (1986).

Swelling power provides evidence of non-covalent bonding between starch molecules; it is affected by factors such as amylose-amylopectin ratio and chain length. Sanchez et al., (2009) reported swelling power values of 0.8 to 15.5 g⁻¹g after investigating starches from 3272 landraces and 772 improved clones in CIAT. Studies conducted by Boakye et al. (2001) on the starch from four varieties of cassava normally cultivated in Ghana, namely: 'Ankra', 'Akosua Tuntum', 'Adwoa Smart' and 'Bosome Nsia' showed that the swelling power ranged between 27.5 and 36.1 g g⁻¹. The swelling power values of starch from 8 cassava genotypes reported by Moorthy (1994) varied from 35.1 to 54.3 g g⁻¹. These reported values indicate that the swelling power figures varied widely. The value obtained from this study ranged from 16.0 – 41.6. These values compared fairly with the values reported by Boakye et al. (2001) and Moorthy (1994), but higher than the values obtained by Sanchez et al. (2009). Sanni et al. (2005) reported that the swelling index of granules reflect the extent of associative forces within the granules, therefore the higher the swelling index, the lower the associative forces.

The significant ($P < 0.05$) differences in treatment means found in swelling power values suggest that the cassava genotypes produce starch with different swelling power properties, that is, the differences in swelling power values can be attributed to varietal differences.

Bainbridge et al. (1996) postulated that, good quality starch will have a low solubility, high swelling volume and high swelling power. High solubility, low swelling volume and swelling power are indicative of poor quality starches that produce thin, low stability when cooked. Using this principle as a yardstick, then all the cassava genotypes, but AFS136 and KW001, produced starches that have good functional properties.

Pasting characteristics

The pasting temperature is the temperature at which irreversible swelling of the starch granules occur leading to peak viscosity. In a constant heating rate experiment as in the case with the Brabender Viscoamylograph, it is directly related to the time to reach peak viscosity. In this experiment the pasting temperatures of starches obtained from the 43 cassava genotypes varied narrowly from 63.3 to 68.2 °C (a range value of 4.9°C).

Pasting temperatures of starch from four varieties of cassava commonly cultivated in Ghana were found to range from 64 to 67 °C (Boakye et al., 2001). In another work by Adomako (2007) on eight cassava genotypes at two locations recorded pasting temperature values of 54.2 to 66.0 °C (a range value of 11.8 °C) and 53.40 to 63.57 °C (a range value of 10.17°C) for Legon and Bunso respectively. Working on seven varieties of a related root and tuber crop, sweet potato, Oduro et al. (2000) observed that the pasting temperatures were relatively high and varied between 72 and 73.3 °C.

The pasting temperatures of starches from seven cassava varieties, namely: M'-4', 'Kalikan', 'H-1687', 'H-2304', 'H-226', 'H-97' and 'H-165' were studied by Moorthy (1994) at the Central Tuber Crops Research Institute, Kerala, India. He found out that starch of varieties 'H-165' and 'H-1687' gelatinized earlier and their pasting temperature range value was relatively higher, that is above 12 °C.

The results reported in this work bear some similarity to the study of Boakye et al. (2001). The pasting temperature values 63.3 to 68.2 °C in this study compare favourably with the value of 64 to 67 °C reported by Boakye and his colleagues. However, the values reported in this work were higher than those recorded by Adomako (2007).

Generally, the higher the amylose, the higher the gelatinization temperature. Amylose molecules, because of their linearity, line up more readily and have more extensive hydrogen bonding and consequently require more energy to break these bonds and gelatinize the starch (Hegenbart, 2009). Based on the findings of Hegenbart accessions 'ADW053', 'AFS001', 'AFS136' and variety 'Bankye Botan' are high in amylose content.

According to Bainbridge et al. (1996) starches with lower pasting temperatures are generally considered to be easier to cook. Therefore, accessions 'OFF086', 'NN42' and 'OFF136' will be easier to cook since they registered the lowest pasting temperatures. However, lower pasting temperatures are also associated with low paste stability, which is usually considered to be an undesirable property. Low pasting temperature and

low paste stability are indicative that fewer associative forces and cross-links are present within the starch granules.

Peak temperature is the temperature at which the starch granule reaches its maximum viscosity. The peak temperature recorded in this study varied from 67.3 to 73.5 °C (a range value of 6.2 °C). Safo-Kantanka and Acquistucci (2002) have reported relatively higher values from 75.5 to 85 °C.

Starch from Bankye Botan recorded the highest paste stability at 95 °C followed by NN42 and AFS136. Paste stability at 95 °C is the difference between paste viscosity value at 95 °C and paste viscosity value after 15 minutes at 95 °C (first holding period). The paste stability at 95 °C measures the tendency of the paste to breakdown during cooking. High paste stability is frequently a requirement for industrial uses of starch (Kim et al., 1995). A starch with low paste stability has very weak cross-linking within the granules and requires less heating. There was significant ($P > 0.05$) difference amongst the paste stability values of the cassava starches.

Paste stability at 50 °C measures the stability of the starch at low temperatures. Most of the starch recorded negative values. This shows that native cassava starches are not suitable for application at low temperatures (Cousidine, 1982).

The peak viscosity is the maximum viscosity reached during the heating phase of the Brabender Visco-Amylograph. At this point, there is a majority of starch granules that are fully swollen but intact. For any

particular type of starch, the more granules that are available to be hydrated the higher the peak viscosity will be.

The peak viscosity values recorded in this study varied from 643.7 to 857.0 Brabender Units (BU). Adomako (2009) reported peak viscosity values that ranged between 830.0 and 988.67 BU, and variation of 1192.33 to 1398.67 BU at Legoon and Bunso respectively. Boakye et al. (2001) reported a range between 445 and 585 BU for starch from four cassava varieties.

The values obtained in this study were relatively lower when compared to the viscosity values reported by Adomanko (2007) but higher than those reported by Boakye et al. (2001). This might be due to due to varietal and environmental differences.

There is significant difference in the peak viscosity of the starch samples ($P < 0.05$). This variation in the peak viscosity might be as a result of the amylose contents of the starches. Oguntunde (1987) reported that the associative bonding of the amylose fraction is responsible for the structure and pasting behaviour of starch granule.

The peak viscosity value and viscosity at 95 °C are measures of the ability of the starch to form a paste on cooking. The higher the viscosity the thicker the paste will be. Jones (1983) and Kim et al. (1995) have noted that a high viscosity is desirable for industrial use for which a high thickening power is required. For this reason, accessions 'AFS136', 'ADW004', 'OFF058' and variety 'Bankye Botan' are the cassava genotypes that can be cultivated to produce starch for industrial purposes

since their starches produced the highest peak viscosities and highest viscosities at 95 °C.

Native starches are not suitable for all starch applications (Cousidine, 1982). Therefore native starch is modified for specific purposes. Starches are modified for a number of reasons. Starches may be modified to increase their stability against excessive heat, acid, shear, time and cooling or freezing; to change their texture; to decrease the viscosity, or to lengthen or shorten gelatinization time. According to Sriroth et al. (2002) for native starch to qualify for modifications it has to possess a minimum peak viscosity of 600 BU. Based on this, starch from 31 out of 43 cassava genotype used in this study could be modified for specific applications.

Viscosity at 95 °C and viscosity 15 min at 95 °C shows how the starch sample can withstand heat. The values obtained in the work shows that as the duration increase at high temperature, the viscosity reduces. This finding agrees with what has been reported in literature (Safokantanka & Acquistucci 2002; Adomako, 2007; Ikegwu et al., 2009; Sanchez 2009). When a starch paste is cooled from the phase at which it was held for 15 minutes at 95 °C to 50 °C, there is an increase in viscosity. This increase indicates the tendency of the starch particles to associate or retrograde.

A low setback or retrogradation viscosity value shows that the starch gives a non-cohesive paste which is useful in many industrial applications as has been reported by Kim et al. (1995). In another work, Sanni et al. (2001) reported that lower set back viscosity during the cooling

of gari indicates higher resistance to retrogradation. In contrast a high setback viscosity value is useful if the starch is to be used for domestic purposes, such as *fufu* and *banku* preparation which require a high viscosity and paste stability at low temperatures (Oduro et al. 2000). According to Nwokocha et al., 2009, starches with high set back viscosity are suitable for use in jelly foods.

Cassava accessions, namely; 'AFS136', 'Bankye Botan', 'KW001', 'KW148' and 'AFS131' produced starch with high setback viscosity values, whilst accessions 'OFF063', 'OFF146', 'Beserebema', 'UCC506', and 'Asaman' produced low setback viscosity.

Based on these observations, therefore, cassava genotypes with low setback viscosity may be recommended for industrial uses and those with high values may be recommended for jelly foods and other domestic purposes.

The breakdown viscosity ranged from 300.5 BU to 659.0 BU. ADW 004 had the highest (659.0 BU). The values of breakdown viscosity of the starch samples were significantly different ($P < 0.05$). Adebowale et al. (2005) reported that the higher the breakdown in viscosity, the lower the ability of the sample to withstand heating and shear stress during cooking. Hence, the starch sample from 'AFS093', 'OFF136', 'ASAMAN' and 'OFF146' might be able to withstand heating and shear stress compared to starch sample from 'ADW004' and 'AFS048' because of their low breakdown value.

Ease of cooking is the maximum duration required to cook starch. The values recorded in this work varied from 1.2 to 5.6 minutes. It is

estimated by subtracting pasting time from peak time. This was similar to what was reported by Sanchez et al. (2009). They reported values ranging from 1.1 to 5.6 minutes with an average of 2.8. According to Ikegwu et al. (2009) short duration for cooking is preferable to save energy cost. There is significant difference ($P < 0.05$) in the ease of cooking of the starch samples.

Functional properties of cassava sweet potato starch mixtures

Swelling volume of starch mixtures of Adehye and sweet potato (Ad:Sp) and that of AFS 048 and sweet potato (AFS:Sp) decrease linearly as the proportion of sweet potato starch increases. However, OFF 146 and sweet potato (OFF:Sp) and Bankye Botan and sweet potato (BB:Sp) did not show any clear pattern. This disparity might be due to varietal difference. The solubility values recorded for all the starch mixtures did not show any regular pattern.

Starch mixtures ((Ad:Sp) and (AFS:Sp)) exhibited linear change in values as the starches were mixed together. Both mixtures recorded linear decrease in swelling power as percentage of sweet potato starch in the sample increases. However, starch mixtures ((OFF:Sp) and (BB:Sp)) did not show any clear pattern. Besides, the value for these mixtures were higher than that of sweet potato values recorded. Daramola and Osanyinlusi (2006) recorded changes in the functional properties of cassava starch when ginger starch was added. Park et al. (2009) studied the pasting properties of potato starch and waxy maize starch mixtures and reported changes in the functional properties of the mixtures.

Solubility values of starch of seven varieties of sweet potato, obtained by Oduro et al. (2000) when they studied the pasting characteristics of these starches are similar to what is presented in this work. They reported a range between 6.82 and 11.94 % which is closely comparable to the value of 12.3% recorded in this work for sweet potato.

Pasting and viscosity characteristics of starch mixtures

Pasting and peak temperature exhibited a linear increase pattern. The pasting and peak temperatures of starch from cassava increased as the proportion of sweet potato in the mixture increased. This result agrees with the finding by Daramola and Osanyinlusi (2006) who recorded increase in pasting and peak temperature of cassava starch when the concentration of ginger increased. According to Hegenbart, (2009) the higher the amylose, the higher the gelatinization temperature. This means that the potato starch might have increased the amylose content of the starch mixture to cause the increase in temperature.

Starch that has been modified to cook at higher temperature can be useful in canning process where gelatinization at elevated temperature will facilitate quick heat penetration while the canned content remains fluid especially in the early heating stages (Daramola & Osanyinlusi, 2006). Based on this, cassava and sweet potato starch mixtures can be exploited in the canning industries.

Ease of cooking was appreciably shorter for OFF:Sp mixtures, while BB:Sp mixtures recorded the highest values. Ease of cooking of cassava starch mixtures was prolonged/enhanced by the addition of sweet

potato starch. It increased as the proportion of sweet potato was increased recording the highest values at 50:50 ratios. However, beyond the 50:50 ratios the duration reduced or shortened.

Peak viscosity, breakdown, set back, paste stability at 95°C and paste stability at 50°C did not show any clear pattern. However, with the exception of C₈Sp₂ (BB:Sp) mixture, the remaining C₈Sp₂ mixtures experienced an increase in paste stability at 50 °C.

Based on the reports on starch mixtures reviewed, the lack of clear cut pattern recorded in this work, for most of the parameters, might be due to the fact that cassava and sweet potato are closely related and might possess similar properties.

A low setback or retrogradation viscosity value shows that the starch gives a non-cohesive paste which is useful in many industrial applications as has been reported by Kim et al. (1995). However, with the exception of AFS048, all the cassava accession recorded low set back viscosity at C₂Sp₈. Based on the findings of Kim et al. (1995), cassava and sweet potato combination of 20:80 ratios could be used to reduce the retrogradation of these starches.

CHAPTER SIX

CONCLUSIONS AND RECOMMENDATIONS

Conclusions

The colour of leaf vein and petiole were predominantly purple, while first leaf colour was mainly light green. A greater number of the cultivars had long petioles and sparse pubescence on young leaves. Stem colour was light brown, storage root surface colour was dark brown while storage root cortex and pulp were white/cream. It seems farmers preferred these morphological traits.

The length and width of the central leaf lobe, and plant height were less variable, thus these descriptive characters may not be useful taxonomically.

Wide variations, however, were found among aboveground matter, first apical branch height, weight of storage root per plant, number of root per plant, fresh root yield, harvest index and number of rotten root per plant. These variations could be used as indicators for selection and breeding to enhance any desirable characteristic. Both qualitative and quantitative morphological traits may serve as valuable tools for preliminary evaluation, since they offer a fast approach for assessing the extent of diversity.

The 43 cassava accessions used were genetically variable and clustered into groups not necessarily based on the source of collection. Molecular markers were more efficient in distinguishing the 43 accessions

into 9 clusters, compared to morphological markers, which grouped them into 4 clusters. Therefore for thorough evaluation, molecular markers should be used for further analysis.

Adult whitefly infestation and mean severity scores of ACMD were high at 6 and 12 WAP, with higher numbers of adult whiteflies found on resistant cassava accessions.

The ACMV was the only CMG strain responsible for the disease symptoms found in this study. The ACMV-1/ACMV-2 primer was more efficient in detecting the virus in the samples in that it detected the virus in more samples than the other primers used.

The capevars variety was the only true resistant genotype identified in this study. The presence of the virus in some of the resistant samples suggests that field resistance, observed as no symptoms, was not necessarily an indication of resistance to virus infection. Therefore, field selection of resistance should be complemented with PCR or inoculation test.

There were significant ($P < 0.05$) differences among the cassava genotypes with respect to number of roots, fresh root weight, fresh root yield and harvest index. There were no significant ($P > 0.05$) differences in starch weight and content per unit weight of tuber, however, starch yield differed significantly ($P < 0.05$) among the genotypes. Thus those genotypes that produced more tubers (kg) had a higher starch yield. Therefore, there is the need to breed for enhanced starch content per unit weight of tuber in order to sustain the starch industry.

Based on percentage starch content, the best 10 genotypes were AFS048, UCC470, ADW053, KW001, KW148, OFF019, OFF145, UCC517, AFS136, and Bankye Botan.

Cassava genotypes least affected by ACMV disease and had higher and desirable agronomic traits, starch content and tuber yield characteristics, were Adehye, ADW 053, Bankye Botan, KW 148, KW 001, Nkabom, OFF 019, ADW 051, Capevars, AFS 041, AFS041, 'UCC517', 'UCC470', 'OFF145', and 'OFF063'. These genotypes could be employed in breeding and selection programmes to produce cultivars with improved characteristics.

Nkabom, ADW051, KW070, OFF 029, KW 181, KW 161, OFF 093, Adehye, AFS027, OFF 145, AFS 126, OFF 058, ADW 063, UCC 506 and OFF023 produced starches with desirable functional properties in terms of low solubility, high swelling volume and swelling power for industrial purposes.

Bankye Botan, OFF 029, DMA 002, AFS 027, OFF 058, ADW 063, AFS 136, ADW 004, KW85, AFS 048, KW 161, Asaman, 'DMA066', 'OFF023', and 'AFS041' produced starch with good pasting characteristics for industrial purposes based on low set back viscosity, high peak viscosity and viscosity at 95 °C.

However, for jelly foods and other domestic purposes, such as the preparation of *fufu* and *banku*. for which lower pasting temperatures and high setback viscosity values are required, AFS136, Bankye Botan, KW 001, KW 148 and AFS 131 were the most suitable even though other accessions such as Adehye and DMA 002 can be used.

Apparently, depending upon what criteria used to select starches for industrial or domestic purposes, the results obtained in this work showed that some of the cassava accessions could be used for both industrial and domestic purposes.

Recommendations

- Eventhough 10 genotypes had potential for industrial starch production; the content per unit tuber weight was low. Hence, selection and breeding programmes should be put in place to develop cultivars with improved starch contents in order to sustain the starch industry.
- Some resistant genotypes haboured the ACMV and could serve as reservoirs for transmission to others. As such, field screening for mosaic resistance should always be complemented with molecular diagnostic tests such as PCR or an inoculation test.
- The Capevars variety, the only resistant genotype identified in this study, may be conserved and used as a parent material for breeding for mosaic resistance.
- The genotypes found to have potential for both industrial and domestic uses should be tested in the various agro-ecological zones of the country to assess the suitability and stability across the country.
- The occurrence of accessions with yellow pulp colour was very low in this study. Hence, it should be conserved to avoid total extinction.

REFERENCES

- Abdullahi, I., Atiri, G. I., Dixon, A. G. O., Winter, S., & Thottapily, G. (2003). Effects of cassava genotype, climate and the *Bemisia tabaci* vector population on the development of African cassava mosaic geminivirus (ACMV). *Acta Agronomica Hungarica*, 51 (1), 37-46.
- Adebowale, Y. A., Adeyemi, I. A., & Oshodi, A. A. (2005). Functional and physicochemical properties of flower of six (*Mucuna* sp). *Afri. J. of Biotech.*, 4(12), 1461-1468.
- Addo-Quaye, A. A., Saah, M. K., Tachie-Menson, C. K. B., Adam, I., Tetteh, J. P., Rockson-Akoly, V. K., & Kitson, J. E. (1993). *Agricultural Science for Senior Secondary Schools, Ministry of Education, Ghana*. India: Gangaram, H and Sons.
- Adomako, W. J. (2007). *Screening of local cassava germplasm for domestic and industrial purpose*. (Unpublished doctoral dissertation). University of Cape Coast, Cape Coast, Ghana.
- Afuakwa, J. J., Asiedu, E. A., Osei, C. K., & Safo-Kantanka, O. (1999). *Varietal characteristics of four improved cassava varieties*. Root and Tuber Improvement Programme (RTIP). Factsheet No.1, Ministry of Food and Agriculture, (MOFA).
- Ahmad, M. (2002). Assessment of genomic diversity among wheat genotypes as determined by simple sequence repeats, *Genome*, 45(4), 646-651.

- Ajay, J., Carlappa, A., & Preme, B. (1999). Evaluation of genetic diversity and genome, fingerprinting of *Pandorea* (Bignoniaceae) by RAPD and inter-SSRPCR. *Genome*, 42(4), 714-719.
- Akinlosotu, T. A. (1985). Studies on the control of the cassava mealybug (*Phenacoccus manihot*) and green spider mite (*Mononychellus tanajoa*) in South Western Nigeria. *Journal of Root Crops*, 9, 33-43.
- Allem, A. C. (1987). *Manihot esculenta* is a native of the Neotropics. *Plant Genetic Resources Newsletter*, 71, 22-24.
- Allem, A. C. (1994). The origin of *Manihot esculenta* Crantz (Euphorbiaceae). *Crop Evol.*, 41, 133-150.
- Alonso-Blanco, C., Peters, A. J., Koornneef, M., Lister, C., Dean, C., van den Bosch, N., ... Kuiper, M. T. (1998). Development of an AFLP based linkage map of Ler, Col and Cvi *Arabidopsis thaliana* ecotypes and construction of a Ler/Cvi recombinant inbred line population. *Plant Journal*, 14, 259-271.
- Alves, A. A. C. (2002). Cassava botany and physiology. In R. J. Hillocks, J. M. Tres & A. C. Bellotti (Eds.), *Cassava: Biology, production and utilization* (pp. 67-89). London: CABI Publishing.
- Amenorpe, E. (2002). *Selection of early bulking and mealy cassava (Manihot esculenta, Crantz) germplasm based on total flesh dry matter, starch and sugar levels and cyanide potential of roots* (Unpublished master's thesis). University of Cape Coast, Cape Coast, Ghana.
- Ampong-Mensah, G. (2000). *Preliminary characterization of cassava germplasm from South-Western Ecozone (Central and Western*

- Region) of Ghana* (Unpublished master's thesis). University of Cape Coast, Cape Coast, Ghana.
- Andrews, D. J., & Kassam, A. H. (1976). The importance of multiple cropping in increasing world food supplies. In R. I. Papendick, P. A. Sfinchez & G. B. Triplett (Eds.), *Multiple cropping* (pp. 1-11). Madison, Wisconsin: Am. Soc. Agron.
- Angel, F., Giraldo, F., Gomez, R., Iglesias, C., Tohme, J., & Roca, W. M. (1992). Use of RFLPs and RAPDs in cassava genome. In W. M. Roca & A. M. Thro (Eds.), *Cassava Biotechnology network* (pp. 62-64) Proceedings of the First International Scientific Meeting of Cassava Biotechnology Network held at Centro Internacional de Agricultura Tropical (CIAT), Cartagena de Indias, Colombia: CIAT
- Annisson, G., & Topping, D. L. (1994). Resistant starch: Chemical structure vs physiological function. *Ann. Rev. Nutr.*, *14*, 297-320.
- Annor-Frimpong, C. (1994). A survey of cassava cultivation practices. In E. Ofori & S. K. Hahn (Eds.), *Tropical root crop for a developing economy* (pp. 216-221). Proceedings of the Ninth Symposium of the International Society for Tropical Root Crops. Accra, Ghana: International Society for Tropical Root Crops.
- Ariyo, O. A., Dixon, A. G. O., & Atiri, G. I. (2005). Whitefly *Bemisia tabaci* (Homoptera: Aleyrodidae) infestation on cassava genotypes grown at different ecozones in Nigeria. *J. Econ. Entomol.* *98*(2), 611-617.

- Asamoah, G.K. (1973). *Soils of the proposed farm site of the University of Cape Coast*. Soil Research Institute (C.S.I.R, Ghana) Technical Report No. 88.
- Asante, I. K., & Offei, S. K., (2003). RAPD-based genetic diversity study of fifty cassava (*Manihot esculenta* Crantz) genotypes. *Euphytica*, 131, 113-119.
- Asaoka, M., Blanshard, J. M. V., & Rickard, J. E. (1992). Effect of cultivar and growth season on the gelatinisation properties of cassava (*Manihot esculenta*) starch. *J. Sci.*, 44, 1366-1368.
- Atwell, W. A., Hood, L. F., Lineback, D. R., Varriano-Marston, E., & Zobel, H. F. (1988). The terminology and methodology associated with basic starch phenomena. *Cereal Foods World*, 33: 306-311.
- Ayanru, D. K. G., & Sharma, V. C. (1982). Effects of cassava mosaic disease on certain leaf parameters of field-grown cassava clones. *Phytopathology*, 72, 1057-1059.
- Bainbridge, Z., TomLins, K., Wellings, K., & Westby, A. (1996). *Methods for assessing quality characteristics of non-grain starch staples (Part 1- Introduction)*. Chatham, U.K: Natural Resource Institute.
- Beck, B. D. A., & Chant, S. R. (1958). A preliminary investigation on the effect of mosaic virus on (*Manihot utilissima*, Pohl) in Nigeria. *Tropical Agric. (Trin.)* 35, 59-64.
- Beeching, J. R., Marmey, P., Gavalda, M-C., Noirot, M., Hayso, H. R., Hughes, M. A., & Charrier, A. (1993). An assessment of genetic diversity within a collection of cassava (*Manihot esculenta*, Crantz)

- germplasm using molecular markers. *Annals of Botany*, 72, 515-520.
- BeMiller, J. N. (1997). Starch modification: Challenges and prospects. *Starch/ Starke*, 49 (2), 127 - 131.
- Bennet ,M. D., Smith J. B., & Heslop-Harrison, J. S. (1992). Nuclear DNA amounts in Angiosperm. *Proc R Soc London Ser B.*, 216, 179-199
- Berry, A., & Kreitman, M. (1993). Molecular analysis of an allozyme cline: Alcohol dehydrogenase in *Drosophila melogaster* on the east coast of North America. *Genetics*, 134, 869–893.
- Bertram, R. B. (1993). *Application of molecular techniques to genetic resources of cassava (Manihot esculenta Crantz, Euphorbiaceae): interspecific evolutionary relationships and intraspecific characterisation* (Unpublished doctoral dissertation). University of Maryland, College Park, Maryland, USA.
- Birador, R. S., Hrishi, N., & Rajendran, P. G. (1978). Genetic variability and correlation studies in cassava (*Manihot esculenta*). *Journal of Root Crops*, 4: 710-715.
- Bligh, H. F. J., Blackhall, N. W., Edwards, K. J., & McClung, A.M. (1999). Using amplified fragment length polymorphisms and simple sequence length polymorphisms to identify cultivars of brown and white milled rice. *Crop Sci.*, 39, 1715-1721.
- Boakye, I. K., Ellis, W. O. Oldham, J. H., & Oduro, I. (2001). Characterization of starch from selected cassava varieties and their possible use for pasta production. *J. Ghana Sci. Association*, 3(2), 51-55.

- Braima, J., Yaninek, J., Neuenschwander, P., Cudjoe, A., Modder, W., Echendu, N., ... Muaka, T. (2000). *Pest control in cassava farms: IPM field guide for extension agent*. Lagos: Wordsmithes Printers.
- Buléon, A., Colonna P., Planchot, V., & Ball, S. (1998). Starch granules: Structure and biosynthesis. *Int J Biol Macromol*, 23, 85–112.
- Carter, S. E, Fresco, L. O, Jones P. G., & Fairbairn, J. N. (1995). Introduction and diffusion of cassava in Agric. IITA Research Guide 49. Training Programme at International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria.
- Carvalho, L. C. B., & Schaal, B. A. (2001). Assessing genetic diversity in the cassava (*Manihot esculenta*, Crantz) germplasm collection in Brazil using PCR-based markers. *Euphytica*, 120, 133-142.
- Chavarriaga-Aguirre, P., Maya, M. M., Tohme, J., Duque, M. C., Iglesias, C., Bonierbale, M. W., ... Kochert, G. (1999). Using micro-satellites, isozymes and AFLPs to evaluate genetic diversity and redundancy in the cassava core collection and to assess the usefulness of DNA-based markers to maintain germplasm collections. *Mol. Breed.*, 5, 263-273.
- Chen-Dao, L., Christian, A. F., Bri, B. S., & Graham, J. S. (2001). Genetic similarities and relationships among cowpea breeding lines and cultivars using microsatellite markers. *Crop Sci.*, 4, 89 -197.
- Chitiyo, M., & Kasele, I. (2004). Evaluation of cassava varieties for yield and adaptability in Zimbabwe. *African Crop Science Journal*, 12(3), 197-203.

- CIAT (Cento International de Agricultural Tropical). (1984). *Morphology of the cassava plant: Study guide*. Cali, Colombia: Centro Internacional de Agricultura Tropical.
- CIAT (Cento International de Agricultural Tropical). (1973). *Annual Report*. Cali, Colombia: CIAT.
- CIAT (Cento International de Agricultural Tropical). (1975). *Annual Report*. Cali, Colombia: CIAT.
- CIAT (Cento International de Agricultural Tropical). (1976). *Annual Report*. Cali, Colombia: CIAT.
- Clerk, G. C. (Ed). (1974). *Crops and their diseases in Ghana*. Tema, Ghana Ghana: Publishing Corporation.
- Ciferri, R. (1938). Wise classification of races of cassava (*Mahihot esculenta* Crantz). Italian Colonial Agricultural Institute, Bullet 40. Florence, Italy.
- Cock, J. H., Franklin, D., Sandoval, G., & Jury, P. (1979). The ideal cassava plant for maximum yield. *Crop Science* 19, 271-270.
- Cock, J. H. 1982. Cassava: A basic energy source in the tropics. *Science*, 218, 755-762.
- Collado, L. S., Mabesa, R. C., & Corke, H. (1999). Genetic variation in the physical properties of sweet potato starch. *Journal of Agricultural Food Chemistry*, 47, 4195-4201.
- Colombo, C., Second, G., & Charrier, A. (2000). Diversity within American cassava germplasm based on RAPD markers. *Genet. Mol. Biol.*, 23(1), 189 -199.

- Colvin, J., Omongo, C. A., Maruthi, M. N., Otim-Nape, G. W., & Thresh J. M. (2004). Dual begomovirus infections and high *Bermsia tabaci* populations: Two factors driving the spread of a cassava mosaic disease pandemic. *Plant Pathology*, 53, 577-584.
- Cosby, P. G., & Raven, J. A. (2007). *Aquatic photosynthesis* (2nd ed.). Princeton: Princeton University Press.
- Cousidine, D. M. (1982). *Foods and food production encyclopedia*. NY: John Wiley Inc.
- Dade, H. A. (1930). *Cassava mosaic*. Accra, Gold Coast Yearbook. Gold Coast: Department of Agriculture.
- Danin-Poleg, Y., Reis, N., Tzuri, G., & Katzir, N. (2001). Development and characterisation of microsatellite markers in Cucumis. *Theor Appl Genet.*, 102, 61–72.
- Danquah, E. Y., Blay, E. T. Bosompem, K. M., & Rosu-Nyarko, J. (2000). Isozyme and storage protein polymorphisms in seventeen accessions of groundnuts (*Arachis hypogaea* L.). *Journal of Ghana Science Association*. 2, 23-30.
- Daramola, B., & Osanyinlusi, S.A. (2006). Investigation on modification of cassava starch using active components of ginger roots (*Zingiber officinale* Roscoe). *African Journal of Biotechnology*, 5, (10), 917-920.
- Dellaporta, S. L., Woods, J., & Hicks, J. B., (1983). A plant DNA mini-preparation: Version II. *Plant Mol. Biol Rep.*, 1, 19-21.

- Delpeuch, F., & Favier, J. C. (1980). Characteristics of starches from tropical food plants: Alpha amylase hydrolysis, swelling and solubility patterns. *Ann. Technol. Agric.*, 29, 53 – 67.
- Dengate, H., & Meredith, P. (1984). Variation in size distribution of starch granules from wheat grain. *Journal of Cereal Science*, 2, 83–90.
- Di Gaspero, G., Peterlunger, E., Testolin, R., Edwards, K. J., & Cipriani, G. (2000). Conservation of microsatellite loci within genus *Vitis*. *Theor. Appl. Genet.*, 101, 301- 308.
- Dimbleby, G. W., & Ucko, P.G (1969). *Domestication and exploitation of plants and animals*. Chicago: Aldine Publishing Co.
- Doku, E. V. (1966). Root crops in Ghana. *Ghana Journal of Science*, 6, 15-36.
- Doku, E. V. (1969). *Cassava in Ghana*. Accra: Ghana University Press.
- Donini, P., Stephenson, P., Bryan, G. J., & Koebner, R. M. D. (1998). The potential of microsatellites for high throughput genetic diversity assessment in wheat and barley. *Genet Res Crop Evol.*, 45, 415–421
- Dorosh, P. (1988). *The economics of root and tuber crops in Africa resources and crop management programme (RCMP)*. Research monograph no. 1. International Institute of Tropical Agriculture, Ibadan, Nigeria: IITA.
- Ekanayake, I. J., Dixon, A. G. O., & Porto, M. C. M. (1996). Performance of various cassava clones in the dry savannah region of Nigeria. In G. T. Kump, M. S. Palaniswamy, V. R. Potty, G. Padmaja, S. Kabeerathumara & S. V. Pillai, (Eds.), *Tropical tuber crops:*

- Problems, prospects and future strategies* (pp. 207-215). New Delhi, India: Oxford & IBH Publishing Company.
- Ekanayake, I. J, Osiru, D. S. O., & Parto, M. C. M. (1997). *Morphology of cassava*. IITA Research Guide 61. Ibadan, Nigeria: IITA
- Ekanayake, I. K. (1995). Cassava in multiple cropping systems: definitions of cropping systems and parameters used. *Tropical Root and Tuber Crops Bulletin*, 8, 3-6.
- Elias, M., McKey, D., Panaud, O., & Anstett, M. C. (2001). Traditional management of cassava morphological and genetic diversity by the Makushi Amerindians (Guyana, South America): Perspectives for on-farm conservation of crop genetic resources. *Euphytica*, 120, 143-157.
- Eliasson, A. C., & Gudmundsson, M. (1996). Starch: Physicochemical and functional aspects. In A.C. Eliasson (Ed.), *Carbohydrates in food* (pp. 431–502). New York: Marcel Dekker.
- Ellis, R. P, Cochrane M. P., Dale M. F. B., Duffus C. M., Lymm, A., Morrison I. M., ... Tiller, S. A. (1998). Starch production and industrial use. *J. Sci. Food Agric.*, 77, 289-311.
- El-Sharkawy, M. A., & Cock, J. H. (1987). Response of cassava to water stress. *Plant Soil*, 100, 345-360.
- El-Sharkawy. M. A. (2003). Cassava biology and physiology. *Plant Molecular Biology*, 56, 481-501.
- El-Sharkawy, M. A., Tafur, S. M., & Cadavid, L. F. (1992). Potential photosynthesis of cassava as affected by growth conditions. *Crop Sci.*, 32, 1336-1342.

- Estessami P, Saunder K, Watt J., & Stanley J (1991). Mutational analysis of complementary sense genes of ACMV DNA-A. *J. Gen. Virol.*, 72, 1005-1012.
- Eun, Y. P., Hyun-Na, K., Jong-Yea, K., & Seung-Taik, L. (2009). Pasting properties of potato starch and waxy maize starch mixtures. *Starch*, 61, 352-357.
- Fargette, D., Fanquet, D., & Thouvenel, J. C. (1988). Yield losses induced by African cassava mosaic virus in relation to the mode and the date of infection. *Trop. Pest Management*, 38, 89-91.
- Fargette, D., Fauquet, C., & Thouvenel, J. C. (1985). Field studies on the spread of African cassava mosaic. *Annals of Applied Biology*, 106, 285-294.
- Fauquet, C., Fargette, D., & Thouvenel, J. C. (1987). *The resistance of cassava to African cassava mosaic*. Yamoussoukro, Cote d'Ivoire: CTA-ORSTOM.
- Fauquet, C. M., & Stanley, J. (2003). Geminivirus classification and nomenclature: progress and problems. *Ann. Appl. Biol.*, 142, 165-189.
- Fernando, D., Javier, M., & Francisco, J. C. (2001). The nucellus degenerates by a process of programmed cell death during the early stages of wheat grain development. *Planta*, 213, 352-360.
- Fondong, V. N., Pita J. S, Rey, M. E.C., de Kochko A., Beachy, R. N., & Fauquet, C. M. (2000). Evidence of synergism between African cassava mosaic virus and a new double-recombinant geminivirus

- infecting cassava in Cameroon. *Journal of General Virology*, 81, 287-97.
- Fontes, E. P. B., Luckow, V. A., & Hanley-Bowdoin, L. (1992). A geminivirus replication protein is a sequence specific DNA binding protein. *Plant Cell*, 4, 597 - 608.
- FAO (Food and Agricultural Organisation of United Nations). (2008). *Facts and figures*. Rome, Italy. Retrieved from www.faostat.org.
- FAO (Food and Agricultural Organisation of United Nations). (2002). *Facts and figures*. Rome, Italy. Retrieved from www.faostat.org.
- FAO (Food and Agriculture Organization). (1995). *Fact and Figures*. Rome, Italy. Retrieved from [http:// www.faostat.org](http://www.faostat.org).
- Freeland-Graves, J. H., & Peckham .G. C. (1987). *Foundations of food preparation* (5th ed.). New York: Macmillan Publishing Company.
- Fregene, M., Suarez, M., Mkumbira, J., Kulembeka, H., Ndedya, E., Kulaya, A., ... Kresovich, S. (2003). Simple sequence repeat marker diversity in cassava landraces: genetic diversity and differentiation in an asexually propagated crop. *Theor. Appl Genet.*, 107, 1083-1093.
- Fregene, M., Angel, F., Gomez, R., Rodriguez, F., Chavarriaga, P., Roca, W., ... Bonierbale, M. (1997). A molecular genetic map of cassava (*Manihot esculenta* Crantz). *Theor. Appl. Genet.*, 95, 431 - 441.
- Fregene, M., Bernal, A., Duque, M., Dixon, A., & Tohme, J. (2000). AFLP analysis of African cassava (*Manihot esculenta*, Crantz) germplasm resistant to the cassava mosaic disease (CMD). *Theor. Appl. Genet.*, 100, 678-685.

- Fregene, M., Okogbenin, E., Mba, C., Angel, F., Suarez, M. C., Guitierrez, J., ... Tohme, J. (2001). Genome mapping in cassava improvement: Challenges, achievements and opportunities. *Euphytica*, 120, 159-165.
- Fregene, M. A., J. Vargas, J., Ikea, F. Angel, J., Tohme, R. A. Asiedu, M. O., ... Roca, W. M. (1994) Variability of chloroplast DNA and nuclear ribosomal DNA in cassava (*Manihot esculenta*, Crantz) and its wild relatives. *Theoretical and Applied Genetics*, 89, 719-727.
- French, D. (1972). Fine structures of starch and its relationship to the organization of starch granules. *Journal of Japanese Society of Starch Science*, 19, 8-25.
- Gallant, D. J., Bouchet, B., & Baldwin, P. M. (1997). Microscopy of starch: Evidence of a new level of granules organization. *Carbohydrate Polymers*, 32, 177-191.
- Garcia, A. M., & Walter, W. M. Jr. (1998). Physicochemical characterization of starch from Peruvian sweet potato selections. *Starch/Starke*, 5, 331-337.
- Geddes, A. M. V. (1990). The relative importance of crop pests in Sub-Saharan Africa. *Natural Resources Institute Bulletin*, 36, 6-12.
- Gepts, P. (1993). The use of molecular and biochemical markers in crop evolution studies. *Evol. Bio.* 27, 15-94.
- Ghislain, M., Spooner, D. M., Rodríguez, F., Villamon, F., Núñez, C., Vásquez, C., ... Bonierbale, M. (2004). Selection of highly informative and user-friendly microsatellites (SSRs) for genotyping

- of cultivated potato. *Theoretical and Applied Genetics*, 108, 881–890.
- Gomez, G. Santos, J., & Validivieso, M. (1984). Cassava Foliage: Chemical Composition, Cyanide Content and Effect of Drying on Cyanide Elimination. *Journal of the Science of Food and Agriculture*. 36, 433-441.
- Goodwin, T. W. (1963). Studies in carotenogenesis. 25. The incorporation of $^{14}\text{CO}_2$, $[2\text{-}^{14}\text{C}]$ acetate and $[2\text{-}^{14}\text{C}]$ mevalonate into b-carotene by illuminated etiolated maize seedlings. *Biochemistry Journal*, 70, 612-617.
- Hahn, S. K., & Keyser, T. (1985). Cassava: A basic food of Africa. *Outlook on Agric.*, 14(2), 95-100.
- Hahn, S. K., Isoba, J. C. G., & Ikotun, T. (1989) Resistance breeding in root and tuber crops at the International Institute of Tropical Agriculture (IITA) Ibadan, Nigeria. *Crop Production*, 8, 147-168.
- Harrison, B. D., Zhou, X., Otim-Nape, G. W., Liu, Y., & Robinson, D. J. (1997). Role of a novel type of double infection in the geminivirus-induced epidemic of severe cassava mosaic in Uganda. *Annals of Applied Biology*, 131, 437-448
- Hearne, C. M., Ghosh, S., & Todd, J. A. (1992). Microsatellites for linkage analysis of genetic traits. *Trends in Genetics*, 8, 288–294.
- Hegenbart, S. (2009). *Understanding starch functionality*. Retrieved from [http:// www.foodproductdesign.com](http://www.foodproductdesign.com).
- Henry, G. W., Andres, W., & Collinson, C. (1998). Global cassava end-uses and markets: Current situation and recommendations for

further study. *A Report of an FAO Consultancy by the European Group on Root, Tuber and Plantain*. Revision 25/10/98, file:/consultancy/FAO/finrep 11.doc.

Hermansson, A. M., & Svegmarm, K. (1996). Developments in the understanding of starch functionality. *Trends in Food Science and Technology*, 7, 345-353.

Hershey, C., Iglesias, C., Iwanaga, M., & Tohme, J. (1994). *Definition of a core collection for cassava*. International crop network series no.10. Rome, Italy: International Plant Genetic Resources Institute.

Hill, B. G. (1968). Occurrence of *Bermisia tabaci* in the field and its relation to the leaf curl disease of tobacco. *South African Journal of Agriculture*, II, 583-594.

Hizukuri, S. (1986). Polmodal distribution of the chain lengths of amylopectins and its significance. *Carbohydrate Research*, 147, 342-347.

Howeler, R. H., & Cadavid, L. F. (1983) Accumulation and distribution of dry matter and nutrients during a 12-month cycle of cassava. *Field Crop Research*, 7, 123-139.

Hudson, R. R., Bailey, K., Skarecky, D., Kwaitowski, J., & Ayala, F. J. (1994). Evidence for positive selection in the superoxide dismutase (sod) region of *Drosophila melanogaster*. *Genetics*, 136, 1329-1340.

Hunt, R. (1990). *Basic growth analysis*. London: Unwin Hym.

IITA (International Institute of Tropical Agriculture). (1976). *Annual Report*. Ibadan, Nigeria: IITA,

- IITA (International Institute of Tropical Agriculture). (1980). *Annual Report*. Ibadan, Nigeria: IITA.
- IITA (International Institute of Tropical Agriculture). (1990). *Annual Reports*, Ibadan, Nigeria: IITA.
- IITA (International Institute of Tropical Agriculture). (2001). *Research guide 55. Physiology of the cassava plant*. Retrived from <http://www.iita.org/info/tm-mat/irgss/irg553.html>.
- Ikegwu, O. J, Nwobasi, V. N., Odoh, M. O., & Oledinma, N. U., (2009). Evaluation of the pasting and some functional properties of starch isolated from some improved cassava varieties in Nigeria. *EJEAFChem*, 8(8), 647-665.
- Imberty, A., Chanzy, H., Peréz, S., Buleon, A., & Tran, V. (1988). The double-helical nature of the crystalline part of A-starch. *Journal of Molecular Biology*, 201, 365-378.
- Imberty, A., & Perez, S. (1988). Conformational analysis and molecular modeling of the branching point of amylopectin. *Int. J. Biol. Macromol.*, 11, 177–185.
- Imberty, A., Buleon, A., Tran, V., & Perez, S. (1991). Recent advances in knowledge of starch structure, *Starch/Starke*, 43, 375-384.
- Irikura, Y., Cock, J. H., & Murray, B. G. (1979). The physiological basis of genotype x temperature interactions in cassava. *Field Crop Research*, 2, 227-239.
- Jacobs, H., & Delcour, J. (1998). Hydrothermal modifications of granular starch with retention of the granular structure: A review. *Journal of Agricultural and Food Chemistry*, 46, 2895–2905.

- Jane, L. J., & Chen, J. F. (1992). Effect of amylose molecular size and amylopectin branch chain length on paste properties of starch. *Cereal Chem.*, 69, 60-65.
- Jangchud, K., Yuthana P., & Vichai, H. (2003). Physicochemical properties of sweet potato flour and starch as affected by blanching and processing. *Starch*, 55, 258-264.
- Janssen, A. (1998). Plants with spider-mite prey attract more predatory mites than clean plants under greenhouse conditions. *Entomol. Exper. Appl.*, 90, 191-198.
- Janssens, M. J. J. (2001). Cassava. In R. H. Raemaekers, (Ed.) *Crop production in tropical Africa*, (pp. 165-187). Brussels, Belgium: Directorate General for International Co-operation.
- Jarne, P., & Lagoda, P. J. L. (1996). Microsatellites, from molecules to populations and back. *Trends Ecology and Evolution*, 11, 424-429.
- Jensen A, K. (2009). *Starch modification*. Retrieved January 16, 2010, from <http://www.kmc.dk/Productsknowhow/Product%20technology/Modifiedstarch/index.as.htm>
- Jones, S. F. (1983). *The world market for starch and starch products with particular reference to cassava (Tapioca)*. Starch Report of the Tropical Development and Research Institute, G173 pp. 13.
- Jones, W.O, (1959). *Manioc in Africa*. California: Stanford University Press.
- Joobeur, T., Perian, N., de Vicente, M. C., King, G. J., & Arus, P. (2000). Development of a second-generation linkage map for almond using RAPD and SSR markers. *Genome*, 43(4), 649-655.

- Kanju, E. (2007, November 26). Hope against cassava brown streak virus. *CTA Spore* 131, 25.
- Kanno, H. A. Dixon. G. O., Asiedu R., & Hahn, S. K. (1991). Host plant resistance of cassava green spider mite (*Mononychellus tanajoa*). In M. N. Alvarez & R. Asiedu (Eds.), *The role of root crops in regional food security and sustainable agriculture* (pp. 103-106). Proceedings of a Southern Africa Regional Workshop Mansa, Zambia. Ibadan: IITA.
- Katayama, K., Komae, K., Kohyama, K., Kato, T., Seiji, T., & Katsumi K. (2002). New sweet potato line having low gelatinization temperature and altered starch structure. *Starch*, 54, 51-57.
- Karp, A., Seberg, O., & Buiatti, M. (1996). Molecular techniques in the assessment of botanical diversity. *Annals of Botany*, 78, 143-149.
- Kaufman, P. B. (1989). *Plant: Their biology and importance*. New York: Harper & Row.
- Kaur, L., Singh, J., & Singh, N. (2006). Effect of cross-linking on some properties of potato (*Solanum terosum* L.) starches. *Journal of the Science of Food and Agriculture*, 86, 1945-1954.
- Kephart ,S. R. (1990). Starch gel electrophoresis of plant isozymes: A comparative analysis of techniques. *American Journal of Botany*, 77, 693-712.
- Ketiku, A. O., & Oyenuga, V. A. (1972). Changes in the carbohydrate constituents of cassava root tuber (*Manihot utissima*, Pohl) during growth. *Journal of the Sci. of Food and Agric.*, 24, 275-278.

- Kim, Y. S., Wiesenborn, D. P., Oir, P. H., & Grant, L. A. (1995). Screening potato starch for novel properties using differential scanning calorimetry. *Journal of Food Science*, *60*, 1060-1065.
- Kizito, E., Bua, A., Fregene, M., Egwang, T., Gullberg, U., & Westerberg, A. (2006). The effect of cassava mosaic disease on the genetic diversity of cassava in Uganda. *Euphytica*, *146*, 45-54.
- Knight, J. W. (1969). *The starch industry*. London: Pergamon Press.
- Kong, L., Dong, J., & Hart, G. E. (2000). Characteristics, linkage-map positions, and allelic differentiation of *Sorghum bicolor* (L.) DNA using simple-sequence repeats (SSRs). *Theor. Appl. Genet.*, *101*, 438-448.
- Kreiger, M., & Ross, K. G. (2002). Identification of a major gene regulating complex social behavior. *Science*, *295*, 328-332.
- Lancaster, P. A., Ingram, J. S., Lim, M. Y., & Coursey, D. G. (1982). Traditional cassava-based foods: Survey of processing techniques. *Econ. Bot.*, *15*(361), 12-45.
- Lian, T. S., & Cock, J. H. (1979). Branching habit as yield determinant in cassava. *Field Crops Res.*, *2*, 281-289.
- Leach, H. W., Mcowen, D. L., & Schock, T. J. (1959). Swelling and Solubility patterns of various starches, structure of starch granule. *Cereal Chem.*, *36*, 53-544.
- Leather, R. J. (1959). *Disease of economic plants in Ghana other than cocoa*. Ghana Ministry of Food and Agriculture, Division Bulletin No 1.

- Legg, J. P., Mallowa, S., & Sseruwagi, P., (2003). First report of physical damage to cassava caused by the whitefly, *Bermisia tabaci* (Gennadius) (Gehiptera: Sternorrhyncha: Aleyrodidae). *Book of abstracts. 3rd international Bermisia workshop* (pp. 23-24). Barcelona: Vertigo Publishers.
- Leuschner, K. (1978). Whiteflies: Biology and transmission of African cassava mosaic disease. In T. Brekelbaum, A. Bellotti & T. C. Lozano (Eds), *Proceedings of the cassava protection workshop* (pp 51-58).Columbia: CIAT.
- Lineback, D. R (1984). The starch granule: Organization and properties. *Bakers Digest*, 58(2), 16.
- Liu, H., & Lelievre, J. (1992). Transitions in frozen gelatinized-starch systems studied by DSC. *Carbohydr. Polym.*, 19, 179-183.
- Liu, K., & Muse, S. V. (2005). PowerMarker: Integrated analysis environment for genetic marker data. *Bioinformatics*, 21(9), 2126-2129.
- Lokko, Y., Danquah, E. Y., Offei, S. K., Dixon, A. G. O., & Gedil, M. A (2005). Molecular markers associated with a new source of resistance to the cassava mosaic disease. *Afr. J. Biotechnol.*, 4(9), 873-881.
- Lozano, J. C., & Booth, R. H. (1994). Diseases of cassava (*Manihot esculenta*, Crantz). *PANS*, 20, 30-54.
- Lundell, C. L. (1939) Plant probably utilized by the old empire, Maya of Peten and adjacent low lands. *Botany*, 24, 37-56.

- Magoon, M. L., Krishnan, R., & Kihakshmi, A. P. (1970). Association of plant and tuber characters with yield of cassava. *Tropical Root and Tuber Crops Newsletter*, 5, 29-30.
- Mahunga, N. M., Yamaguchi, Y., Almazan, A. M., & Hahn, S. K. (1987). Reduction of cyanide during processing of cassava to some traditional African foods. *Journal of Food and Agriculture*, 1, 11 - 15.
- Manners, D. J. (1968). Studies on carbohydrate-metabolizing enzymes. Sweet-corn branching enzymes. *Carbohyd. Res.*, 8, 72-81.
- Maravalhas, N. (1984). Carotenoids de *Manihot esculentus* Crantz. *Brazil Instituto Nacional de Pesquisas de Amazonia Publicacao*, 6, 33-38
- Marmey, P., Beeching, J. R., Hamon, S., & Charrier, A. (1994). Evaluation of cassava (*Manihot esculenta*, Crantz) germplasm using RAPD markers. *Euphytica*, 74, 203-209.
- Mason, R. R. (1956). Cassava varieties in Fiji. *Fiji Agric Journal*, 27, 88-93.
- Mat-Hashim, D. G., Moorthy, S. N., Mitchell, J. R., Hill, S. E., Linfoot, K. J., & Blanshard, J. M. V. (1992). The effect of low levels of antioxidants on the swelling and solubility of cassava starch. *Starch/Starke*, 44, 471-475.
- Matsuoka, Y., Mitchell, S. E., Kresovich, S., Goodman, M., & Doebley, J. (2002). Microsatellites in *Zea*-variability, patterns of mutations, and use for evolutionary studies. *Theoretical and Applied Genetics*, 104, 436-450.

- May, B. (1992). Starch gel electrophoresis of allozymes. In A. R Hoelzel, (Ed.), *Molecular genetic analysis of populations: A practical approach* (pp. 141-148). Oxford: Oxford University Press.
- Mba, R. E. C, Stephenson P, Edwards K, Melzer S, Mkumbira J, Gullberg U., ... Fregene, M. (2000). Simple sequence repeats (SSR) markers survey of the cassava (*Manihot esculenta* Crantz) genome: Towards an SSR-based molecular genetic map of cassava. *Theor Appl Genet.*, 102, 21-31.
- McDowell. I., & Oduro, K. A. (1983). Investigation of the β - carotene content of the yellow varieties of cassava (*Manihot esculenta* Crantz). *J. Plant Foods*, 5, 169-171.
- MOFA (Ministry of Food Agriculture). (2000). Root tuber. *A Newsletter of the Ghana root and tuber improvement MOFA*, p. 2.
- MOFA (Ministry of Food and Agriculture). (2005). *Agriculture in Ghana, facts and figures*. Accra; PPME, MOFA.
- Montilla, J., Castillo, P., & Wiedenhofer, H. (1975). Efecto de la incorporacion de havina de yucca a morga en racionres parapollos de engrarde. *Agron. Trop.*, 25, 259-266.
- Moorthy, S. N. (1994). *Tuber crop starches*. Kerala, India: Central Tuber Crops Research Institute.
- Moorthy, S.N., & T. Ramanujam, (1986). Variation in the properties of starch in cassava varieties in relation to age of the crop. *Starch/Starke*, 38, 58-61.
- Moorthy, S. N. (2001). Behaviour of cassava starch in various solvents. *Starch/Starke*, 37, 372-374.

- Morgante, M., & Olivieri, A. M. (1993). PCR-amplified microsatellites as markers in plant genetics. *Plant Journal* 3, 175–182.
- Morgante, M., Hanafey, H., & Powell, W. (2002). Microsatellites are preferentially associated with nonrepetitive DNA in plant genome. *Nature Genetics* 30, 194–200.
- Mound, L. A (1965). Effect of leaf hair on cotton whitefly populations in the Sudan Ge zira. *Empire Cotton Growing Review*, 42, 33-40.
- Moyib, O. K., Odunola, O. A., & Dixon A. G. O. (2007). SSR markers reveal genetic variation between improved cassava cultivars and landraces within a collection of Nigerian cassava germplasm. *African Journal of Biotechnology*, 6(23), 2666-2674.
- Msikita, W., Braima, J., Nnodu, E., Legg, J., Wydra, K., & Ogbe, F. (2000). *Disease control in cassava farms: IPM field guide for extension agents*. Lagos: Wordmithes.
- Munsell Color. (1977). *Munsell color charts for plant tissues*. Maryland, USA: Macbeth Division of Kollmorgen Corporation.
- Mutsaers, H. J. W., Ezuma, H. C., & Osiru, D. S. O. (1993). Cassava based intercropping. *A Review of Field Crop Research*, 34, 431-437.
- Nasser, N. M. (1978). Conservation of the genetic resources of cassava. (*Manihot esculenta*, Crantz): Determination of wild species location with emphasis on probable origin. *Economy Botany*, 32, 311-318.
- Nei, M. (1983). Estimation of genetic distances and phylogenetic trees from DNA analysis. *Proc. 5th World Cong. Genet. Appl. Livestock Prod.*, 21, 405-412.

- Nowjee, N. C. (2004). *Melt processing and foaming of starch*. Cambridge: University of Cambridge Press.
- Ntawuruhunga, P., Whyte, J. B. A., & Rubaihay, O. (1995). Correlation and principal components studies of some plants attributes at early plant growth in cassava to assist breeding work in predicting potential high yielding genotypes. *African Crop Science Conference Proceedings*, 6, 119-125.
- Nweke, F. (2004). *New challenges in cassava transformation in Nigeria and Ghana*. Washington, USA: International Food Policy Research Institute.
- Nweke, I., Dixon, A. G. O., Asiedu, R., & Folayan, S. A. (1994). *Cassava varietal needs of farmers and the potential for production growth in Africa*. Ibadan, Nigeria: IITA.
- Nwokocha, L. M., Aviara, N. A., Senan, C., & Williams, P. A. (2009). A comparative study of some properties of cassava (*Manihot esculenta*, Crantz) and cocoyam (*Colocasia esculenta*, Linn) starches. *Carbohydrate Polymers*, 76, 362–367.
- Obanni, M., & BeMiller J. N. (1997). Properties of some starch blends. *Cereal Chem.*, 74, 431-436.
- Odemerho, F.O., & Avwunudiogba, A. (1993). The effects of changing cassava management practices on soil loss: A Nigerian example. *The Geographical Journal*, 159, 66-69.
- Odongo, B., & Orone, J. (1992). Predation of cassava green mite by exotic and indigenous predatory mites in Nawulongge, Uganda: Preliminary studies In M. O. Akoroda & O. B. Arene (Eds.), *Tropical root*

- crops: Promotion of root crop based industries. Proceedings of the fourth Triennial Symposium of the International Society for Tropical Root Crops Africa* (pp. 36-48). Ibadan, Nigeria: IITA.
- Oduro, K. A., Yankson, M. F., & Appiah, J. (1989). Importance and vegetative variations in the little-known yellow cassava in Africa. In T. Tuner (Ed.), *Discovery and innovation* (pp. 41-49). Nairobi, Kenya: Academy Science Publishers.
- Oduro, I., Ellis, W. O., Aryeetey, S. K., Ahenkora, K., & Otoo, J. A. (2000). Pasting characteristics of starch from new varieties of sweet potato. *Trop. Sci.*, 40, 25-28.
- Offei, S. K., Owuna-Kwakye, P., & Thottappilly G. (1999). First Report of East african cassava mosaic begomovirus in Ghana. *The American Phytopathological Society Journal*, 56, 187-193.
- Ogbe, F. O., Nnodu, E. C., & Odurukwe, S. O. (1996). Control of African cassava mosaic disease incidence and severity. *Tropical Science*, 36, 174-181.
- Ogbe, F. O., Atiri, G. I., Robinson, D., Winter, S., Dixon, A. O., Quin, F. M., & Thottappilly, G. (1999). First report of East African cassava mosaic begomovirus in Nigeria. *Plant Dis.* 83, 398-402.
- Ogbe, F.O. (2001). *Survey of cassava begomoviruses in Nigeria and the response of resistant cassava genotypes to African cassava mosaic begomovirus infection* (Unpublished doctoral dissertation). University of Ibadan, Ibadan, Nigeria.

- Ogunbodede, B. A. (1997). Multivariate analysis of genetic diversity in Kenaf (*Hibiscus cannabinus*). *Africa Crop Science Journal*, 5(2), 127-133.
- Oguntunde, A. O. (1987). Review: Starch modification for food application in Nigerian. *Food Journal*, 5, 102 – 107.
- O'Hai, S. K. (1989). *Cassava*. Florida: University of Florida press.
- Okeke. J. E. (1979). Evaluation of agronomic traits in six Nigeria varieties on representative ultisol and alfisol of Southern Nigeria. *Journal of Root Crops*, 5(2), 25-31.
- Okogbenin, E., Marin, J., & Fregene, M. (2006). An SSR-based molecular genetic map of cassava. *Euphytica*, 147(3), 433-440.
- Okoli, P. S. O., & Wilson, G. F. (1986). Response of cassava (*Manihot esculenta* Crantz) to shade under field conditions. *Field Crops Research*, 14, 349-360.
- Omongo, C. A. (2003). *Cassava whitefly, Bemisia tabaci, behaviour and ecology in relation to the spread of the cassava mosaic pandemic in Uganda* (Unpublished doctoral dissertation). University of Greenwich, UK.
- Onwueme, I. C. (1978). *The tropical tuber crops - yam, cassava, sweet potatoes and cocoyams*. Chichester: John Wiley and Sons.
- Onwueme, I. C. (1982). *The tropical tuber crops: Yams, cassava, sweet potatoes and cocoyams*. New York: John Wiley and Sons Ltd.
- Oostergetel, O. G. T., & Van Bruggen, E. F. J. (1993). The crystalline domains in potato starch granules are arranged in a Helical Fashion. *Carbohydrate Polymers*, 21, 9-12.

- Oslén, K. M., & Schaal, B. A. (1999). Evidence of the origin of cassava: Phylogeography of *Manihot esculenta*. *Academy of Sciences of the United States of America*, 96, 10, 5586-5591.
- Otim, M., Legg, J., Kyamanywa, S., Polaszek, A., & Gerling, D. (2005). Population dynamics of *Bemisia tabaci* (Homoptera: Aleyrodidae) parasitoids on cassava mosaic disease-resistant and susceptible varieties. *Biocontrol Science and Technology*, 16(2), 205-214.
- Otim-Nape, G. W., Shaw, M. W., & Thresh, J. M. (1994). The effects of African cassava mosaic gemini virus on the growth and yield of cassava in Uganda. *Trop. Sci.* 34, 43-54.
- Oyetunji, O. J., Ekanayake, I. J., & Osonubi, O. (1998). Photosynthetic photochemical response of cassava (*Manihot esculenta* Crantz) to water stress and mycorrhizal inoculation. *Tropical Agriculture (Trinidad)*, 75(2), 328-329.
- Park, E., Kim, H., Kim, J., & Lim, S. (2009). Pasting properties of potato starch and waxy maize starch mixtures. *Starch-Starke*, 61, 205-214.
- Parker, G. D., K. J. Chalmers, Rathjen, A. J., & Langridge, P. (1998). Mapping loci associated with flour colour in wheat (*Triticum aestivum* L.). *Theoretical and Applied Genetics* 97, 238-245.
- Parkes, E. (2009). *Using molecular markers to identify new sources of CMD resistant genes for cassava improvement*. 2nd GSS Workshop, CIAT, Cali, Colombia.
- Perera, L., Russell, J. R., Provan, J., & Powell, W. (2000). Use of microsatellite DNA markers to investigate the level of genetic

diversity and population genetic structure of coconut (*Cocos nucifera* L.). *Genome* 43(1), 15-21.

- Pérez, J. C., Morante, N., López, J., Lenis, J. I., Jaramillo, G., Ceballos, H., & Calle, F. (2001). Advantages of the new cassava breeding scheme at CIAT. In N. J Taylor, F. Ogbe & C. M. Fauquet (Eds.), *Abstract book for the Fifth International Scientific Meeting of the Cassava Biotechnology Network* (pp. 98-101). Missouri, USA: Donald Danforth Plant Science Centre St. Louis.
- Perring, T. M. (2001). The *Bemisia tabaci* species complex. *Crop Protection*, 20, 725 -737.
- Pineda, M. J., & Rubio, R. R. (1972). Un concepo Nuevo en el levantar de novillas para ganaderia de leche, *Rev. ICA* (7), 405-413.
- Pita, J. S., Fondong, V. N., Sangaré, A., Otim-Nape, G. W., Ogwal, S., & Fauquet, C. M. (2001). Recombination, pseudorecombination and synergism of geminiviruses are determinant keys to the epidemic of severe cassava mosaic disease in Uganda. *Journal of General Virology*, 82, 655-65.
- Purseglove, J. W. (1987). *Tropical Crops: Dicotyledons*. London: Longman.
- Quaye, S. K. (1979). *The cyanide content of the leaves and tubers of different varieties of cassava*. A research project report submitted to the Department of Nutrition and Food Science, University of Ghana. 45pp.
- Queller, D. C., Strassmann, J. E., & Hughes, C. R. (1993). Microsatellites and kinship. *Trends in Ecology and Evolution*, 8, 285–288.

- Radley, J. A. (1976). *Industrial uses of starch and its derivatives*. Essex, England: Applied Science Publishers Ltd.
- Raghu, D., Senthil, N., Saraswathi, T., Raveendran, M., Gnanam R., Wnkatachalam, R., ... Mohan, C. (2007). Morphological and simple sequence repeats (SSR) based finger printing of South Indian cassava germplasm. *International Journal of Integrative Biology*, 2, 141-149.
- Ramanujam, T., Nair, G. M., & Indira, P. (1984). Growth and development of cassava (*Manihot esculenta* Crantz) genotypes under shade in a coconut garden. *Turrialba*, 34, 267-274.
- Ramanujam, T. (1990). Effect of moisture stress on photosynthesis and productivity of cassava. *Photosynthetica*, 24, 217-224.
- Reichel-Dolmantoff, G. (1957). Momil: A formative sequence from the Sinu Valle, Colombia. *Am Antig.*, 22, 226-234
- Renvoize, B. S. (1972). The Area of origin of *Manihot esculenta* as a crop plant: A review of the evidence. *Economy Botany* 26, 352-360.
- Rickard, J. E, Asaoka, M., & Blashard, J. M. V. (1991). The pysioco-chemical properties of cassava starch. *Tropical Science*, 31, 189-207.
- Ring, S. N. (1993). Functional behaviour of starches. *Nutition and Food Science*, 5, 20-23.
- Roa, A. C., Maya, M. M., Duque, M. C., Tohme, J., Allem, A. C., & Bonierbale, M. W. (1997). AFLP analysis of relationships among cassava and other *Manihot* species. *Theoretical and Applied Genetics* 95, 741-750.

- Rogers, D. J. (1963). Studies on *Manihot esculenta* Crantz cassava and related species. *Bull Torrey Bot Club*, 9, 43-54.
- Rogers, D. J. (1965). Some botanical and ethnological considerations of *Manihot esculenta*. *Economy*, 19, 369-376.
- Rogers, D. J., & Appan, H. S. (1973). A monograph of *Manihot esculenta* with an explanation of the taximetrics method used. *Econ. Bot.*, 27, 1-13.
- Saal, B., & Wricke, G. (1999). Development of simple sequence repeat markers in rye (*Secale cereal* L.). *Genome*, 42, 964-972.
- Safo-Kantanka, O., & Acquistucci, R. (2002). The role of common salt in maintaining hot-paste viscosity of cassava starch. In D. Duffour, G. M. O'Brien & R. Bes, (Eds.), *Cassava flour and starch: Progress in research and development* (pp. 1- 6). Colombia: CIAT.
- Sajilata, M. G., Rekha S., Kulkarni, P. (2006). Resistant starch: A review. *Comp. Rev. Food Sci.*, 5, 1-17
- Sanchez, T., Salcedo, E., Ceballos, H., Duffour, D., Mafia, G., Morante, N., ... Moreno, I. (2009). Screening of starch quality traits in cassava (*Manihot esculenta* Crantz). *Starch/Starke*, 61, 12-19.
- Sanni, L., Dixon B. M, Akanya J, Okoro C. I, Alaya Y, Egwuonwu C. V., ... Dixon A. (2005). *Standards for cassava products and guidelines for export*. Ibadan, Nigeria: IITA.
- Sauer, J. D. (1993) *Historical Geography of crop plants: A select roster*. Boca Raton, Florida: CRC Press.

- Schierwater, B., & Ender, A. (1993). Different thermostable DNA polymerases may apply to different RAPD products. *Nucleic Acids Research*, 21, 4647–4648.
- Schlötterer, C. (2004). The evolution of molecular markers: Just a matter of fashion? *Genetics*, 5(1), 63-69.
- Shannon, J. C., & Garwood, D.L. (1984). Genetics and physiology of starch development. In R. L. Whistler (Ed.) *Starch: Chemistry and technology* (pp. 25-87). NY: Academic Press.
- Smith, J. S. C., & Smith, O. S. (1989). The description and assessment of distances between inbred lines of maize: The utility of morphological, biochemical and genetic descriptors and a scheme for testing of distinctiveness between inbred lines. *Maydica*, 34, 151-161.
- Smith, J. S. C., Kresovich, S., Hopkins, M. S., Mitchell, S. E., Dean, R. E., Woodman., ... Porter, K. (2000). Genetic diversity among elite sorghum inbred lines assessed with simple sequence repeats. *Crop Sci.*, 40, 226-232.
- Sneath, P. H. A., & Sokal, R. R. (1973). *Numerical taxonomy: The principles and practice of numerical classification*. San Francisco: Freeman.
- Spooner, D., R. van Treuren, P., & de Vicente M. C. (2005). *Molecular markers for genebank management*. IPGRI Technical Bulletin No. 10. International Plant Genetic Resources Institute, Rome, Italy. Retrieved:http://www.biodiversityinternational.org/Publications/pubfile.asp?ID_PUB=1082. 126 p.

- Sriroth, K., Piyachomwan, K., Sangseethong, K., & Oates, C. (2002). Modification of cassava starch. In *Proceedings of 10th International Starch Convention on Cassava Starch, Cracow, Poland, 11-14 June, 2002 (pp. 103-112)*. Cali, Columbia: CIAT.
- Sserubombwe, W. S., Thresh, J. M., Otim-Nape, G. W., & Osiru, D. S. O. (2001). Progress of cassava mosaic virus disease and whitefly vector populations in single and mixed stands of four cassava varieties grown under epidemic conditions in Uganda. *Ann. Appl. Biol.* 135, 161-170.
- Stewart, W. D. P. (1974). Nitrogen fixation by photosynthetic microorganisms. *Ann. Res. Microbiol.*, 27, 283–316.
- Storey, H.H., & Nichols, R.F.W. (1938). Studies on the mosaic of cassava. *Ann. Appl. Biol.*, 25, 790-806.
- Stute, R., & Kern, H. (1994). *U.S. Patent No, 5,324,532*. Washington, D.C.: U.S. Patent and Trademark Office.
- Tams, S. H., Bauer, E., Oettler, G., & Melchinger, A. E. (2004). Genetic diversity in European winter Triticale determined with SSR markers and co-ancestry coefficient. *Theor. Appl. Gene*, 108, 1385 – 1391.
- Terry, E. R., & Hahn, S. K. (1990). The effect of cassava mosaic disease on growth and yield of a local and an improved variety of cassava. *Tropical Pest Management*, 26, 34 – 37.
- Thresh, J. M., Otim-Nape, G. W., & Nichols, R. F. W. (1994). Strategies for controlling African cassava mosaic geminivirus. *Adv. Dis. Vector Res.*, 10, 215-236.

- Thresh, J. M., Otim-Nape, G. W., Legg J. P., & Fargette, D. (1997). African cassava mosaic virus disease: The magnitude of the problem. *African Journal of Root and Tuber Crops*, 2, 13-19.
- Thomas, D. J., & Atwell, W. A. (1998). *Starches*. St. Paul, MN: Eagan Press.
- Thottapilly, G., Crouch, J. H., & Quin, F. M. (1996). Overview of DNA marker Research at IITA. DNA marker-assisted improvement of the staple crops of the sub-Saharan Africa. In H. C. Jonathan & T. Abdou (Eds.), *Proceedings of the workshop on DNA markers* (pp.109-118). Ibadan, Nigeria: IITA.
- Tonukari, N. J., Thottapilly, G. Q., & Mignouna, H. D. (1997). Genetic polymorphism of cassava within the Republic of Benin detected with RAPD Markers. *Afr. J. Crop Sci.*, 5(93), 219-228.
- Tsou, S. C. S., & T. L. Hong. (1992). The nutrition and utilization of sweet potato. In W. A. Hill, C. K. Bonsi & P. A. Loretan (Eds.), *Sweet potato technology for the twenty-first Century* (pp. 221-232). Tuskegee: Tuskegee University Press.
- Ugent, D., Pozorski, S., & Pozorki, T. (1986). Archaeological manioc (*Manihot esculenta*) from coastal Peru. *Econ.Bot.* 40, 78-102.
- Ugorji, N. (1998). *Genetic characterization of cassava cultivars in Nigeria: Morphological and molecular markers*. (Unpublished master's thesis). University of Ibadan, Ibadan, Nigeria.
- Veltkamp, H. J. (1985). *Physiological causes of yield variation in cassava (Manihot esculenta, Crantz)*. (Unpublished doctoral dissertation). Agricultural University, Wageningen, Netherlands.

- Weising, K., Nybom, H., Wolff, K., & Kahl, G. (2005). *DNA Fingerprinting in plant Principles, methods, and applications*. (2nd ed.). New York: CRC Press.
- Wheatley, C. C., & Chuzel, G. (1993). Cassava: The nature of the tuber and use as a raw material. In R. Macrae, R. K. Robinson & M. J. Sadler (Eds.), *Encyclopaedia of food science, food technology and nutrition* (pp. 734-743). San Diego, CA: Academic Press.
- Whistler, R., BeMiller, J., & Paschall, E. (1984). *Starch: Chemistry and technology* (2nd ed.). New York: New York Academic Press.
- Whitkus, R., Doebley J., & Wendel, J. F. (1994). Nuclear DNA markers in systematics and evolution. In R. L. Phillips & I. K. Vasil (Eds.), *DNA-based markers in plants*, (pp. 116–141). Dordrecht: Kluwer Academic Publishers.
- Wholey, D. W., & J. H. Cock. (1974). Onset and rate of bulking in cassava. *Experimental Agric.*, 10, 193-198.
- Yan-Ping, G., De-Ping, G., Jian-Ping, Z., Hui, L., Yan, P., Qiao-Mei, W., ... Gui-Zhen, R. (2004). Photosynthetic rate and chlorophyll fluorescence in leaves of stem mustard (*Brassica juncea* var *tsatai*) after turnip mosaic virus infection. *Plant Science*, 168, 57-63.
- Young, W. P., Schupp, J. M., & Keim, P. (1999). DNA methylation and AFLP marker distribution in the soybean genome. *Theoretical and Applied Genetics*, 99, 785–792.
- Yu, J. K., Mangot, J., Thompson, L., Edwards, K. J., Slabaugh, M. B., & Knapp, S. J. (2000). Allelic diversity of simple sequence repeats

among elite inbred lines of cultivated sunflower. *Genome*, 45, 652-660.

Zaidul, I. S. M., Norulaini, N. A. N., Omar, A. K. M., Yamauchi, H., & Noda, T. (2007). RVA analysis of mixtures of wheat flour and potato, sweet potato, yam and cassava starches. *Carbohydrate Polymers*, 69, 784–791.

Zhou, X., Liu, Y., Calvert, L., Munoz, C., Otim-Nape, G.W., Robinson, D.J., & Harrison, B. D. (1997). Evidence that DNA-A of a geminivirus associated with severe cassava mosaic disease in Uganda has arisen by interspecific recombination. *J. Gen. Virol.*, 78, 2101-2111.

Zobel, H. F. (1988). Starch crystal transformations and their industrial importance. *Starch/Sarke*, 40, 1-12.

