NUMERICAL TAXONOMIC STUDIES OF THE EAST AFRICAN HIGHLAND BANANAS (*Musa* AAA-East Africa) IN UGANDA ● Deborah A. Karamura



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NUMERICAL TAXONOMIC STUDIES OF THE EAST AFRICAN HIGHLAND BANANAS (*Musa* AAA-East Africa) IN UGANDA

Deborah A. Karamura

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ABSTRACT

The purpose of this study was to determine the variation pattern that exists in the East African Highland bananas (*Musa* AAA) grown in Uganda, estimate levels of dissimilarity caused by different growing conditions, and establish a flexible provisional classification and identification system. Techniques of numerical taxonomy were employed to determine the variation pattern and these included two different coefficients, three different clustering methods, principal component analysis and classificatory discriminant analysis. Sixty one morphological characters were employed to determine differences among the 238 accessions available for the study; 192 accessions were from the national banana germplasm collections, 46 were from farmers' fields in selected sites. Phenetic classifications resulting from different analyses were compared with an independent subjective classification. The phenetic classifications agreed with the subjective classification with regard to the positions of the majority of accessions. Accessions which were inconsistently placed in the cluster analyses were classified by classificatory discriminant analysis and 84 clones were identified. It was advantageous to compare different methods because they often gave complementary results. For example the comparison of cluster analysis versus principal component analysis revealed similar clusters of accessions in the phenograms and along the first four principal components.

The East African Highland bananas have been kept as a subgroup within a Group (*Musa* AAA) as defined by the International Code of Nomenclature for Cultivated Plants. The smallest distinguishable units in the subgroup are clones and they have been grouped into clone sets. In summary four categories have been adopted in the classification of the East African Highland bananas; Group, subgroup, clone set and clone. Five clone sets were delineated, based mainly on qualitative characters. The clone sets were distinct enough for new accessions to be fitted into them. Beer and Musakala were very distinct from each other and from the rest of the clone sets. Nakabululu, Nfuuka and Nakitembe were least distinct from each other. These data can now be used to develop hypotheses concerning the evolutionary background of the East African Highland bananas and to exploit vigour, pest and disease resistance in the representative clones of the different clone sets proposed.

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To my husband, Eldad Karamura,
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Chapter 1

GENERAL INTRODUCTION

1.1 Background

Bananas are among the most important food crops worldwide (Samson, 1992). They include diverse types such as dessert, cooking, roasting and beer bananas. These types are based on use of their end products (Acland, 1971). Dessert bananas are those bananas consumed raw at ripeness and are usually distinguished by the sweet flavour of the fresh fruit when ripe. Cooking bananas are consumed when cooked and in much of the world they have always been referred to as plantains (Swennen & Vuylsteke, 1987). However, there are two types of cooking bananas. The first type is cooked when the fruits are green and provides a starchy staple nutritionally similar to the potato (Simmonds, 1966). Bananas of this type can be allowed to ripen and then eaten as dessert bananas. For this reason they cannot be called true plantains. The second type of cooking banana is unpalatable raw even when the fruits are ripe and therefore requires cooking before being consumed. These are the true plantains (Swennen & Vuylsteke, 1987). The beer bananas are bananas whose pulp is bitter and astringent. They can be eaten neither raw nor cooked. However, juice and alcohol can be made from this type of bananas, hence the name.

Bananas are also scientifically referred to by their genome groupings. The crop encompasses a range of diploids, triploids and tetraploids. These are categorised into genome groups on the basis of their ploidy levels and the genomes which they contain. Simmonds and Shepherd (1955) suggested that edible bananas originated from two wild and seedy species, *Musa acuminata* Colla (2n=22), and *Musa balbisiana* Colla (2n=22) which are native to Southeast Asia, resulting in a series of diploid, triploid and tetraploid bananas. The resulting genome groups were classified as AA, AB, AAA, AAB, ABB, AABB, AAAB, ABBB with the letters A and B representing the contributions of *M. acuminata* and *M. balbisiana* respectively. Table 1.1 gives a few examples of cultivars with their genome group and use.

World banana production has been estimated at 80.6 million tonnes (FAO, 1994) of which only 15% is exported. The dessert banana is a major export crop of the Caribbean countries where the Cavendish bananas have accounted for 10% of the world's production of bananas and plantains (Daniells, 1990).

The remaining bananas are consumed in the producing countries where it is estimated that one half of the production is eaten raw as dessert fruits and the other half cooked to provide a starchy staple (INIBAP, 1994). Bananas constitute the fourth most important

starchy staple after cassava, sweet potatoes and yams (Table 1.2). It is estimated that over 70 million people worldwide subsist on this crop (INIBAP, 1994). For many countries the banana is an important import substitution crop without which the producer countries would have to import food. In Africa, plantains are cultivated from the lowlands of Guinea and Liberia to the central basin of Zaire (Devos *et al.*, 1978) where 20% of the world's plantains are grown (FAO, 1994). In East Africa the cooking bananas alone account for 54.4% of the total world output of bananas, and the region is the major consumer of the banana staple as well (INIBAP, 1994).

Table 1.1: Genome groups and uses of some widely grown Ugandan bananas

Genome group	Cultivar name	End product use
AAA	Bogoya Omweru (Gros Michel)	dessert
	Nakitembe	cooking
	Kabula	beer
AAB	Nakatansese	cooking/roasting
ABB	Kivuvu	cooking
	Kayinja	beer
AB	Kisubi	beer
	Sukali Ndiizi	dessert

Table 1.2: Production levels of some major starchy staples (million metric tonnes) FAO, 1994

Region	Cassava	Yam	Sweet potato	Cooking bananas
Africa	72.8	29.1	6.9	20.7
Asia	48.4	0.2	1.1	0.8
South America	30.1	0.3	1.2	5.6
Central America	1.0	0.4	114.3	1.6
World total	152.3	30.1	124.5	29.7

1.2 Bananas in East Africa

Bananas are the staple food in many of the higher altitude, wetter areas of East Africa (Fig. 1.1). They are mostly grown as a subsistence crop. However, there is much internal trading, often over long distances from the growing areas to the main town markets. No banana export trade has developed. Among the reasons for this are the distance between the important growing areas and the sea, the distance between East Africa and the major consuming areas, i.e. Europe and U.S.A, and the fact that most of the export varieties selected for high rainfall, hot, humid conditions give poor results in the main growing areas of East Africa, which are cooler and drier (Acland, 1971).

In Uganda the crop is a staple food in all parts which do not experience a pronounced dry season, i.e. the area within about 80km of the shore of Lake Victoria, the south western highlands, the slopes of Mt. Elgon in the east and the well watered areas of the western part of the country (Fig.1.1).

In Tanzania, bananas are a staple food in the high rainfall areas, located in higher altitudes and low lying places around Lake Victoria in addition to valley bottoms,



Figure 1.1: Principal East African Highland banana growing regions (shaded black).

where they are only a minor constituent of the local diet (Acland, 1971; Ngeze, 1994). In Kenya, bananas are widely grown in the western part, in areas surrounding Lake Victoria, on the slopes of Mt. Kenya and along the coast. Maize, however, is the staple food in these areas. Bananas are also dominant features in the landscapes of Rwanda, Burundi, and eastern Zaire (Baker & Simmonds, 1951; Sebasigari, 1987) (Fig.1.1).

The East African bananas, whose names are too numerous to catalogue, have evolved to suit the local environment. They can be grouped into three categories: dessert bananas, plantains and the East African Highland (cooking and beer) bananas. East Africa to the west and north of Lake Victoria is regarded as a secondary diversity centre for bananas (Simmonds, 1966).

The dessert bananas include *Musa* AB and AA diploids and one AAB triploid (Prata) but the majority are AAA triploids. The dessert bananas together with the plantains, are found growing in backyards and in fields around villages in East Africa (De Langhe *et al.*, 1994). Depending on the clone, the East African Highland bananas are used for boiling and cooking or in beverage preparations. The former are called cooking bananas, the latter beer bananas (Sebasigari, 1987).

The cultivation of bananas has become woven into the socioeconomic life of the communities in Eastern Africa. The crop is a key component in both the food security and the agricultural sustainability of the region. It has an extended harvest period which ensures food and income throughout the year. In addition farmers in the tropics can intercrop bananas with legumes and can feed animals on by-products (peels and pseudostems) of the crops. The plant has been used for medicinal purposes (Wainwright, 1953), for celebrating marriage and for other rituals (Howes, 1928; Price, 1994). Virtually all components of the plant have found use in the homesteads, and many domestic industries like making baskets, carpets, shoes and a host of indoor decorations have been developed (Wainwright, 1953; Price, 1994).

The Highland bananas reduce soil erosion on steep slopes and are principal sources of mulch for maintaining and improving soil fertility (INIBAP, 1986). This is because the giant herb (2-8 metres high) with its large leaves easily creates closed canopies which assist in arresting rain impaction and direct insolation, both of which are important in soil conservation (Karamura, 1992). The leaves as well as stems rot as they are being broken down by micro-organisms which gives good aeration to the soil and this also adds organic matter to the soil.

Environmentally, the banana is “a tropical forest “ because once established, it enters a phase of continuous growth. Gardens as old as 50-60 years used to be known in Uganda (Tothill, 1940) but with the current problems of the crop, these are rare.

1.3 Current problems of the banana crop in Uganda

Uganda has a long tradition of banana cultivation dating back to the 13th century (Dale, 1955). In almost all aspects (acreage, annual production, employment prospects, environmental conservation, medicinal purposes and food security), bananas of the East African Highland type are by far the most important food crop in Uganda. They constitute 85% of the bananas grown, the introduced beer clones (*Musa* AB, ABB) constitute 11%, the dessert type constitutes 3% while the plantains constitute only 1% (Karamura *et al.*,

1996). However, during the 1970s and 1980s, Uganda witnessed drastic yield declines (Fig. 1.2) in traditional growing areas of the east and central regions of Uganda (MOA, 1991).

Such yield declines have led to replacement of cooking bananas with exotic beer bananas (seen as more hardy) causing an increase in hectarage and/or annual crops (maize, sweet potatoes, cassava) (Gold *et al.*, 1993; Zake, 1992). It has been suggested that yield decline of the Highland bananas resulted from a combination of factors including population pressures, socio-economic considerations (including labour and competing activities), declining soil fertility, pest outbreaks (weevils, nematodes) and introduction of new pests, particularly a nematode (*Radopholus similis* (Cobb) Thorne) and a fungal disease (*Mycosphaerella fijiensis* Morelet) (Gold *et al.*, 1993). Reduction in yield of the crop and farmers moving away from growing the crop created considerable concern within the Ministry of Agriculture. In 1989, the Ministry developed a National Banana Research Programme (NBRP) to address the issue of the banana decline. However, as late as 1990 there were no baseline data to document regional production levels, farming systems, production constraints, cultivar distribution and germplasm available in the country. Moreover, there was no systematic or coherent approach to banana research. Therefore, with the support of the Rockefeller Foundation and the International Development Research Council, a collaborative program was developed between the NBRP, Makerere University and the International Institute of Tropical Agriculture (IITA) to a) characterise Ugandan banana-based cropping systems b) determine principal production constraints and c) prioritize research needs and directions.

Germplasm characterisation was one of the prioritized research needs. There was an urgent need to search for what germplasm is available in the country, where and how it is distributed and maintained, and whether any clones are resistant to the pests and diseases mentioned above. The germplasm in the country had not yet been characterised, nor evaluated for its potential use against the above constraints.

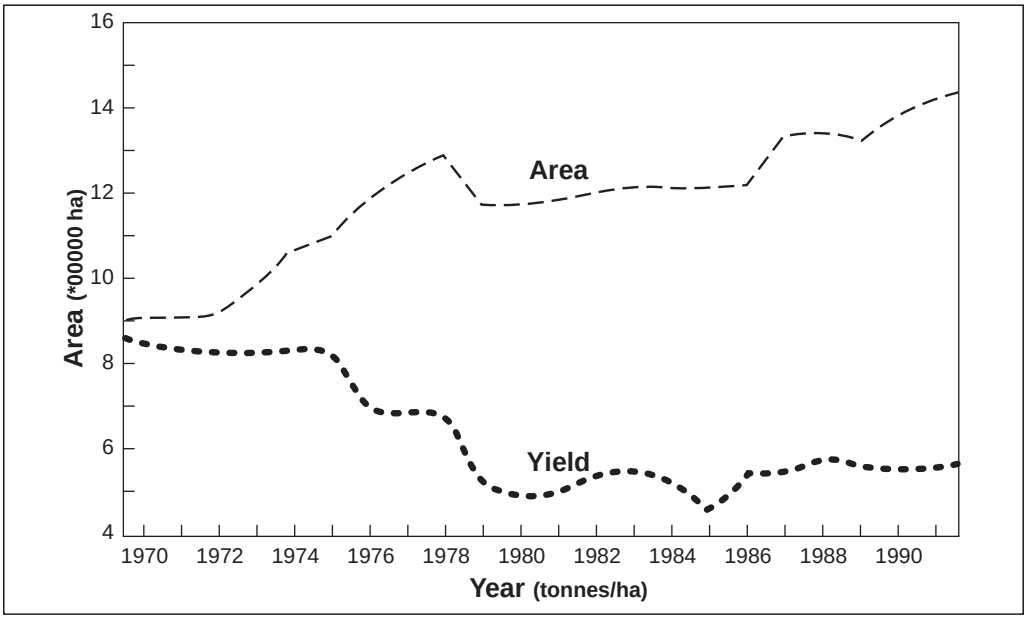


Figure 1.2: Banana production in Uganda between 1970-1990 (Ministry of Agriculture Report, Uganda 1991).

1.4 Banana germplasm collections

The Highland bananas are seedless like all other cultivated bananas and in East Africa propagation of the crop has been exclusively by vegetative means. This means that germplasm collections (which were already established but incomplete and under severe threat of diseases and pests) had to be maintained as either field collections or *in vitro* because these are the conservation methods which cater for vegetatively propagated plants. Both methods are expensive and each has certain associated risks. The cultural conditions for *in vitro* conservation vary and depend on the one hand on the requirements of the plant species concerned and on the other hand on the technical and spatial possibilities available. Efforts are always necessary to develop improved methods. Risks in *in vitro* conservation arise from somaclonal variation which is frequent and the need for frequent regeneration.

Field collections on the other hand are disadvantageous compared to *in vitro* ones in that they occupy a larger space and they are exposed to damage by different environmental factors like diseases, pests, wind and drought and this makes them more expensive to maintain (Brown, 1995). Banana field germplasm collections have been preferred over *in vitro* ones because they are immediately available for demonstration, study and use. They also do not need regeneration as frequently as *in vitro* ones.

Uganda has two major banana germplasm collections: one at Kabanyolo Agricultural Research Institute, under the mandate of Makerere University, which is used mainly for training purposes, and one at Kawanda Agricultural Research Institute, managed by the National Agricultural Research Organization, which is used to study variation, provide material for breeding, and evaluate pest and disease resistance. Together the two collections contain more than four hundred accessions, representing all major genome groups but mainly East African Highland bananas (*Musa* AAA-East Africa).

The collections however need to be studied to assess how completely they represent the range of diversity of bananas in the country, to set priorities for further collection, and to reduce duplication, if any, within each collection. This is because there has not been any systematic collection of new accessions or evaluation of accessions in these collections since their establishment.

Banana germplasm in Uganda offers challenges in that, since there is no test for clonal identity, many clones may have been collected several times under different names because there is a long list of different vernacular names attached to each clone (Kyobe, 1981; Karamura & Karamura, 1994). This is one of the principal problems facing researchers, farmers, and extensionists alike. Some names may be synonyms due to the many languages spoken, others may just refer to environmental effects on the phenotype of the crop or to somatic mutations. It is very difficult to know how many clones there are in the country. Shepherd (1957) put their number at fifty one. McMaster (1962a) estimated the number to be between forty and forty-five different clones.

It is generally believed that the first Highland banana introduced in Uganda gave rise to all present day Highland bananas through mutation (Tothill, 1970). If this is true, there are thus old mutations and new ones. Clones claimed by farmers to be the oldest on their farms or in their villages have several variants. Some of these variants may appear to be generated by the environment but majority are most probably mutants. For example ‘Nakabalulu’ or ‘Embururu’ is a widely cultivated clone and also considered as one of the oldest clones in Uganda by the farmers. It has several different variants. One variant has a fruit pulp which is astringent and bitter, hence used for beer making.

It is called ‘Nakabululu Embidde’ in the central region or ‘Enshenyuka’ in the Western region. Another variant has a fruit pulp which is insipid and not bitter, hence used for cooking when still green. This is the original ‘Nakabululu’, known all over the country. A third variant has vegetative parts (stem and leaves) which are red, not green, but the fruits are still used for cooking. This is ‘Nakabululu Omumyufu’ in the central region or ‘Nakasabira’ or ‘Mukite’ in the Eastern region. There are many other names applied to these variants of ‘Nakabululu’ as one moves across the banana growing regions.

Neither the farmers in the next village, nor researchers, extensionists or the international community know the synonyms among these many names. Yet, scientists need to use the same name for the same genotype to enable them to publish results which can be compared with results obtained by others working on the same crop. The description of clones by farmers lacks precision although it can illustrate how local farmers identify the clones found in their areas. The farmers are always sure of the identity of clones in their own gardens, but the identity of clones outside their gardens is always a problem. Lack of clear clone identity in the crop has resulted in unnecessary duplication with regard to collection, conservation and research.

1.5 Research objectives

From the foregoing, the following problems have been observed. There is a general lack of a systematic and comprehensive study of the Highland banana germplasm. This is because, since their introduction to East Africa, the Highland bananas have diversified through somatic mutations and have acquired names in various local languages so that determining their synonyms is difficult. Classification and identification have been further complicated by the fact that many morphological characters are greatly influenced by the growing conditions, some mutate readily yet the majority are not so sharply discontinuous.

Determination of synonyms soon became essential. The clones and their names are too many to be dealt with individually as farmers do, they need to be classified. But then there are no standardized charts or tables of these names to explain the synonyms within the crop. There is also no field or easily comprehensible key for identifying the clones, making the situation worse. It is therefore necessary to develop identification schemes for these clones. The synonyms need to be documented, to reduce confusion. The folk taxonomies of farmers need to be collected and put together as an important ethnobotanical link to the systematic studies of the clones (van der Maesen, 1988) . Based on this background therefore, the following objectives were proposed:

(1) to determine the variation pattern existing within the East African Highland banana clones of Uganda and estimate levels of dissimilarity caused by different growing conditions. This would estimate the amount of variation existing and enable the description of accessions in the national banana germplasm collections

(2) to classify the Highland bananas into identifiable and manageable groups for ease of reference and communication purposes by using different methods of multivariate analyses and to assess the relative merits of these analyses.

(3) to identify the most useful characters in grouping the Highland bananas and develop a provisional identification system for them. This would facilitate the identification of accessions and provide possible synonyms in the crop.

Chapter 2

EVOLUTIONARY AND TAXONOMIC BACKGROUND OF BANANAS

2.1 Banana classification and distribution

Bananas belong to the order Zingiberales and family Musaceae. Members of this family are large herbs 2-9 metres tall with an aerial trunk consisting of compacted leaf sheaths which grow directly from the top of the corm (Lawrence, 1951; Purseglove, 1972). Musaceae contains only two genera, *Musa* and *Ensete* (Simmonds, 1966; Cobley & Steele, 1976). The genus *Ensete* is distributed in a wild state in Africa from the Cameroons throughout East Africa down to Transvaal in South Africa. A few species are also found from northeast India to the Philippines and New Guinea (Purseglove, 1972). The genus differs from *Musa* by being monocarpic, non-suckering with a distinctively swollen base, and having large-sized seeds while *Musa* produces suckers and has small seeds (Cobley & Steele, 1976; Samson, 1992). The genus *Musa* contains 30-40 species and all wild species are diploids (2n = 2x = 14, 18, 20, 22) and native to South East Asia (Stover & Simmonds, 1987). The genus *Musa* is divided into five series, based mainly on the basic chromosome numbers, orientation and arrangement of flowers in the inflorescence. The series are *Musa*, *Rhodochlamys*, *Callimusa*, *Australimusa* and *Ingentimusa* (Argent, 1976; Simmonds and Weatherup, 1990a). Series *Musa* is the largest with 13-15 species, the most diversified and considered the most ancient (Purseglove, 1972). It is widely distributed, extending from Southern India to Japan and Samoa (Purseglove, 1972). Series *Musa* has the basic chromosome number of eleven and a pendent or semi-pendent inflorescence (Cheesman, 1948; Simmonds, 1966). This series includes the dessert bananas, cooking bananas and plantains now grown throughout the tropics. The wild species of series *Musa* can reproduce both sexually and asexually (by suckers from a corm). Among the fifteen wild species are *Musa acuminata* and *M. balbisiana* both of which have contributed to the origin of the majority of edible bananas (Purseglove, 1972; Stover & Simmonds, 1987). Another species implicated in the evolution of cultigens is *M. schizocarpa* Simmonds. *M. schizocarpa* may have been involved in the origin of some of the diploids cultivated in Papua New Guinea (Sharrock, 1990). *M. acuminata* originates in Malaysia and is very variable, containing 7-8 subspecies. Four of the subspecies overlap in the Malesian centre of diversity while others form disjunct populations on islands far removed from the main area of distribution. The four subspecies which overlap are ssp. *malaccensis* Simmonds, ssp. *microcarpa*

Simmonds, ssp. *burmannica* Simmonds, and ssp. *siamea* Simmonds. Among the disjunct subspecies is ssp. *banksii* (F. Muell.) Simmonds from Papua New Guinea which is morphologically more distinct than all the other subspecies and at times treated as a different species. *Musa balbisiana* on the other hand originates in the drier parts of India and is widely distributed from there to the Philippines and New Guinea but absent in central Malaysia. It is hardier and more drought and disease-resistant than *M. acuminata* (Simmonds, 1966), but not as variable as M. *acuminata*. No subspecies of *M. balbisiana* have been described. Other known species of series *Musa* are listed in Table 2.1.

Table 2.1: Classification of the genus *Musa* after De Langhe (1969).

Series	Species	2n
Musa	*M. acuminata* Colla	22
	ssp. *malaccensis* Simmonds	
	ssp. *microcarpa* Simmonds	
	ssp. *burmannica* Simmonds	
	ssp. *burmanniccoides* De Langhe & Devreux	
	ssp. *siamea* Simmonds	
	ssp. *banksii* (F.Muell.) Simmonds	
	ssp. *errans* Allen	
	ssp. *zebrina* nom.nud.	
	M. flaviflora Simmonds	
	M. balbisiana Colla	
	M. itinerans Cheesman	
	M. basjoo Siebold	
	M. schizocarpa Simmonds	
	M. nagensium Prain	
	M. sikkimensis Kurz	
	M. cheesmani Simmonds	
	M. ochracea Shepherd	
	M. truncata (Ridl.) Shepherd	
Rhodochlamys	*M. ornata* Roxb. + 3 other species	22
Australimusa	*M. textilis* Nees	20
	M. maclayi F.Muell. + 3 other species	
Callimusa	*M. coccinea* Andr.+ 3 other species	20
Ingentimusa	*M. ingens* Simmonds	14, 18

2.2 Evolution of cultivated bananas

The first and probably most crucial step in the evolution of edible bananas was the development and, subsequently, the selection of parthenocarpy and seed sterility in *Musa acuminata*, giving rise to the edible diploid (AA) cultivars. The wild seedy diploid *M. acuminata* has fruits inedible by human beings but which can be eaten by birds, bats and monkeys. Edible bananas evolved from the seedy wild types by becoming able to produce parthenocarpic fruits, which are seedless so long as the female flowers are not pollinated. Female sterility developed later so that fruits from pollinated flowers were also seedless (Simmonds, 1962). Edibility was improved greatly by parthenocarpy and seed sterility.

It is believed that by the process of chromosome restitution at meiosis in diploid edible *M. acuminata*, there arose AAA (*acuminata*) triploids which now dominate the world's bananas. Due to the wide variability of *M. acuminata*, it is not clear which subspecies were involved in the evolution of edible bananas. However, variation in anthocyanin pigments among the edible diploids suggested that three different subspecies could have been involved. These were ssp. *malaccensis*, ssp. *banksii* and ssp. *zebrina* (Horry & Jay, 1990). The triploids were more vigorous and had bigger fruits than the diploids. Different edible diploids and triploids of *acuminata* spread in cultivation and some reached India.

There has never been a natural edible parthenocarpic diploid (BB) form of *M. balbisiana* nor triploids of this species (BBB) (Simmonds, 1966; Simmonds & Weatherup, 1990b; Jarret, 1990; Simmonds, 1995). Two cultivated bananas, Cardaba and Saba, from the Philippines were treated as BBB types by Vakili (1967) and later Valmayor *et al.* (1981), but recent isozyme studies suggested that Cardaba could be an ABB (Jarret & Litz, 1986) while the scoring technique of Simmonds (1990) put both Saba and Cardaba in ABB. Further isozyme studies may be necessary to confirm that Saba is an ABB.

Another important step in the evolution of bananas was the crossing of AA (and perhaps AAA too) cultivated bananas with the wild *M. balbisiana* (BB). As *M. acuminata* derivatives spread, extending their range in the territories of the wild seeded *M. balbisiana*, natural hybridization occurred resulting in several genome combinations. Hybridization probably took place repeatedly as cultivation of edible triploids of *M. acuminata* continued to spread into the territory of the wild diploid *M. balbisiana*. Armed with good edibility (imparted by the diploid AA) and dry environment tolerance (imparted by the diploid BB), the resulting hybrids (AB, AAB and ABB) extended their geographical ranges out from the wetter tropics into the seasonally drier zones. At present there are many clones of cultivated bananas in existence, belonging to different genome groups, of which AA, AAA, AAB and ABB are the most numerous.

2.3 Introduction of bananas to Africa

The time of introduction and route of bananas from their presumed native centre of diversity in South East Asia to Africa remain a subject of speculation. It is however very important to understand how these introductions occurred, as a basis for understanding the phenotypic variability now found in the continent. It is also important to know the types of banana currently found in Africa to be able to explain their probable introductions to the continent.

There are three categories of bananas found in Africa today. The first category is the cultivars of the East African coast and the nearby islands. These are bananas of different genome groups including edible AA, various AAA, AB and ABB. The AB and ABB are considered to be of recent introduction both in inland Africa and along the coast (Baker & Simmonds, 1951, 1952; Shepherd, 1957; De Langhe *et al.*, 1994). The coastal cultivars are limited in number (per genome group) and no genome group dominates the landscape neither is any of them a staple along the coast (De Langhe *et al.*, 1994). The African plantains (AAB) constitute the second category. They are mainly found in the humid forest lowlands of tropical central and western Africa. The East African Highland bananas (AAA-EA) grown on the East African plateau are the third category. These are cooking and beer cultivars different from the *M. acuminata*

triploids found along the East African coast, which are mainly dessert types. The East African Highland bananas are extensively pigmented black or brown with glossy pseudostems and robust dirty green leaves deeply split along the veins. The coastal AAA triploids have bright coloured pseudostems and leaves which are not deeply split along the veins. For quite a long time no Highland bananas have been reported to occur along the East African coast. Together with the African plantains, the East African Highland bananas are believed to have increased in number and diversity through somatic mutations. They are said to be endemic to the regions where they are found (Shepherd, 1957; Simmonds, 1959). The East African Highland bananas and the African plantains have a traditional dominance in the agricultural systems of tribes who cultivate them and they are staple foods in their respective regions. The Highland bananas are boiled or fermented into beer while the plantains are roasted. They are grown in different ways too: the Highland bananas are grown in groves or plantations which can last for decades while the plantains are grown in backyard gardens which do not last for decades. Their high level of diversity and utilization in their respective areas indicate that they have been cultivated there for a long time.

Differences in genome groups, distribution and utilization by different groups of people are evidence to suggest that these bananas did not come to Africa through the same route or at the same time. It is assumed that plantains originated in southern India (Simmonds, 1966) while triploid *M. acuminata* originated in the Malesian area. It was from these areas that these bananas came to Africa probably occupying suitable ecological conditions along their routes and being influenced by various human cultures.

Modern views about the coming of bananas to Africa can be linked to the general view of the trade relations and contacts that existed between the different Asian regions (e.g between India and Indonesia) or between Asia and the coast of East Africa. These contacts include the migration of Indonesian people to Madagascar between 0-500 AD and the Arab trade and influence after 600 AD along the coast of East Africa (Simmonds, 1966; Purseglove, 1972). These contacts produced a number of crop dispersals mostly from Asia to Africa (Schoenbrun, 1993; Reynolds, 1927). It is therefore possible that bananas (mainly the AA and AAA) could have reached the East African coast during these movements and this is a widely accepted view.

Other views suggest that bananas moved from India through the Holy Land (Jerusalem) or Saudi Arabia to Egypt where bananas were unknown before 650 A.D. It is believed that there was some Arab influence over trade around the Mediterranean region and bananas could have been introduced to these places in that way. It appears that after the introduction of bananas to Egypt, the people there liked them and went ahead to plant them. Banana was also considered a holy plant in the Koran, and had spread to Tunisia and Spain by the 12th and 13th centuries (Wainwright 1952; Price, 1994). From Egypt banana plants might have been transported to central Africa through the valleys of the river Nile (Vansina, 1984). However movement of bananas from India through the Holy Land or Saudi Arabia to Africa is unlikely because the climatic conditions along these routes are too dry for the crop and not humid enough for even the bananas with a B genome to survive there (Purseglove, 1972). Thus dispersal by way of Middle Eastern countries and the Horn of Africa is not widely supported.

It was stated earlier that bananas along the coast are of different genome groups which include the edible AA and different AAA genotypes. It can be assumed that an edible AA or AAA was carried to inland East Africa and it evolved to produce the current diversity found there. It is believed that the moment bananas were introduced in inland

East Africa by traders along the coast, they were quickly adopted by inland tribes who spread them further inland (Baker & Simmonds, 1951; Thomas, 1955; Simmonds, 1959; Kirkman, 1959). Others are of the view that Indonesians were also able to enter inland Africa to take the bananas there due to various routes of trade which had developed. This is supported by the dhows and canoes which were copied from the Indonesians by the Bantu tribes inside Africa (Roscoe, 1922).

The introduction of bananas from the coast to inland Africa is further supported by evidence from languages. The general word for bananas in Uganda is Tooke (plural, Matooke; the plant being a Kitooke). This word recurs with minor alterations in a broad corridor stretching through western Tanzania, northern Malawi and down the Ruvuma valley near the coast in Tanzania. This distribution of the word seem to indicate the route of matooke from the coast and probably from Madagascar to inland East Africa (Johnston, 1922). Yet, on either side of the corridor there are quite different words which are not even related to Tooke. Tooke indicates the Highland bananas whereas the kobo, kova, konjwa and gonja found on either sides of the Tooke corridor are related to plantains. It is as though Tooke introduction came and cut across an already existing area of banana cultivation (Wrigley, 1989; Rossel, 1990). However, no language studies have been carried out in Madagascar to work out the origin of the word “Tooke”.

The crucial questions are why are there absolutely no Highland bananas in regions near and along the coast and how did the crop become so central in the Lake Victoria region? According to the Baganda tribe in the central part of Uganda, the Highland banana was introduced into Uganda (and subsequently to other countries of the Great Lakes region) by Kintu, their tribal ancestor. According to this legend, Kintu brought the banana from an area near Mount Elgon in the North-East of Uganda about 1000 A.D. This is an area outside Buganda, the native land of the Baganda. It is believed by the Baganda that the banana plant which was brought by Kintu was planted at Magonga in Busujju county in current Mubende district, and all other bananas grown in Buganda originated from that banana plant (Wainwright, 1952; McMaster, 1962b). The same legend is held by Banyoro and Batoro tribes to west of the Baganda zone and the Bagisu in the eastern part of Uganda (in the Elgon). The Bagisu (Bantu tribe in the eastern part of Uganda) believe that bananas originated from the hills (the Elgon) and that it came with the earliest Bamasaba (the first Bagisu to arrive in the area).

History connected with these Bantu tribal migrations to East Africa indicates that they came from regions around the sources of the rivers Zaire and Zambezi, and another region around where river Benue and river Niger join. They then moved into East Africa and to the rest of Africa. The people they found in the region were hunters, gatherers and fishermen. The Bantu people were farmers and they practised mixed cropping. Their original food included yam (*Dioscorea* species), *Cucurbita* species, a type of bean and oil palm (*Elaeis* species). They had developed skills in making and using various implements and they kept on adding attractive food plants as they became available. These could have included bananas (De Langhe *et al.*, 1994). As soon as bananas were introduced to them, they found them attractive and started planting them. However, by the time the Bantu migrations to East Africa took place (between A.D. 500-1000) (Murdock, 1976) bananas had just been introduced to the Western part of Africa.

It is possible that an AAA banana was introduced to the Lake Victoria region and through mutation it diversified. East Africa is a land of contrast both in its physical features and socially so that the different climate, the physical nature of East Africa and the social background of the region played part in the diversification of the different clones.

2.4 Taxonomic studies of *Musa* species

The evolution of edible bananas provides important background information for our understanding of the taxonomy of bananas particularly how to treat the various sorts already discussed. Several facts are worth considering before going deep into the explanation of the taxonomy of this crop. It is now clear that two wild seeded diploid species were involved in the ancestry of edible bananas and plantains of section *Musa*. Some edible bananas have been derived entirely from one of the species and occur as edible diploids, triploids or tetraploids. Other edible bananas arose as diploid, triploid or tetraploid hybrids between the two basic wild species. The triploid and tetraploid hybrids contain different numbers of sets of chromosomes from each of the two species. In discussing the taxonomy and nomenclature of bananas, the first problem to be addressed is how many species should be recognised in the above complex and what they should be called. The complex can be sorted into three groups by phenotype: those that appear to be derived from just *M. acuminata*; those that appear to be derived from *M. balbisiana* only and finally those that appear to have both wild species in their ancestry.

2.4.1 Status of the wild diploids

The two wild diploid bananas are morphologically distinct (Table 2.2). The A genome diploid is very variable with 7-8 subspecies. The B genome diploid is less variable but phenetically distinct from the A genome diploid.

Table 2.2: Morphological differences between *M. acuminata* and *M. balbisiana*

Characters	*M. acuminata*	*M. balbisiana*
Pseudostem colour	brown or black blotched	green-yellow
Petiole canal margins	spreading	incurved
Peduncle	hairy	glabrous
Inflorescence orientation	oblique to pendulous	subhorizontal to oblique
Pedicels	short	long
Fruits	dark green and non-waxy	pale green and waxy
Ovules	two rows of ovules in each loculus	four rows of ovules in each loculus
Male bud	slightly waxy, ovoid, bracts convolute	waxy, broadly ovate, bracts imbricate
Bract colour	bright red to deep violet outside, light red to yellow inside	various shades , of purple outside scarlet inside
Male flower colour	cream, yellow or orange with no pink flush	cream with a strong pink flush
Compound tepal	same length as free tepal	twice as long as free tepal

The two wild diploids are therefore very different morphological species despite their ability to hybridise, as others have noted (Cheesman, 1948b).

2.4.2 Diploid edible bananas (AA)

Most edible diploid bananas evolved entirely from wild A genome diploid by becoming seedless (Simmonds, 1966). Since the A genome diploid is a variable species with several different subspecies (Simmonds & Weatherup, 1990a), the different edible diploids may have originated independently and more than once. This has been confirmed through flavonoid studies (Horry & Jay, 1988). The question arises as to whether these edible diploids should be treated as a different species from their wild parent. Apart from being seedless the edible diploids remain within the morphological range of their wild parent and therefore should be treated as the same species as their wild parent (Bell, 1969; Pickersgill, 1986a; Trehane *et al.*, 1995; Robinson, 1996).

There are no edible diploids derived from *M. balbisiana* (Simmonds, 1966) but if there were, they too would be treated as belonging to the same species as their wild progenitor.

2.4.3 Triploid edible bananas (AAA)

Edible triploids of *M. acuminata* arose from edible diploids which produced unreduced female gametes (Simmonds, 1966). When fertilised by a normal haploid male gamete, these gave rise to triploid edible bananas. Again these triploids may have originated independently many times, possibly with contributions from the different subspecies of *M. acuminata*. This is supported by molecular data on differences in DNA of 9 diploid and triploid accessions of bananas (Howell *et al.*, 1994). A similar question arises as to whether the triploids too should belong to the same species as the wild and cultivated A diploids. Increase in the number of chromosomes by autopolyploidy will cause very little change in the observable appearance of the plant although there may be increased vigour in different parts of the plant (Jeffrey, 1986b). This is because autopolyploidy does not add anything new in the way of genetic material and no special taxonomic significance has been attached to the condition since morphological variation will usually be within the limits of the normal variation pattern of the species concerned (Bell, 1969; Pickersgill, 1986a). These AAA bananas could therefore be treated as the same species as the AA diploid. Similarly, BBB bananas would be of the same species as both wild and cultivated *M. balbisiana*.

2.4.4 Interspecific hybrids

The hybrids which evolved from the two wild species include bananas with the AB, AAB and ABB genomes. ABBB, AAAB and AABB tetraploid hybrids are also presumed to occur in South-East Asia (Richardson *et al.*, 1965). There are naturally occurring hybrids as well as those which have been raised in cultivation. The triploids may have arisen in two ways; when an unreduced female gamete of an A genome diploid banana is fertilised by a haploid male gamete from the B genome diploid banana or when an unreduced female gamete of an interspecific diploid hybrid is fertilised by a haploid male gamete from an A genome diploid banana (Pickersgill, 1995). This will give rise to AAB triploids. The ABB triploids may be produced when an unreduced female gamete of an interspecific diploid hybrid is fertilised by a haploid male gamete from a B genome diploid banana. Triploid bananas may also produce unreduced female gametes and when fertilised by a haploid male gamete they give rise to tetraploids. The resulting interspecific hybrids will be different from the autopolyploids since they will consist of two different genomes. Most edible bananas are hybrids. According to the morphological species concept and without knowledge of their origin, they would be regarded as another species differing from those derived entirely from each of the two wild species.

2.4.5 Names for the wild diploids and the hybrid complex

So far in the above complex three possible morphological species can be observed: those derived from each of the two wild species and those which are hybrids. Having decided to recognise three species, the next question is to find out their correct names from the names available within the genus *Musa*. Names become available through being validly published and their application is decided by their type specimens.

According to the International Code of Botanical Nomenclature, a taxon can have only one scientific name applied to it and by which it has to be known. The application of this name is determined by means of its nomenclatural type. The type is an element on which the description associated with the original publication of the name was based or considered to have been based. The term "element" means different things according to the rank of the taxon. For example the type of the name of a species is a herbarium specimen or plates of the plant from which the original description validating the name was drawn. The type of the name of a genus is the species on which the description validating the name was based. A name will therefore apply to the taxon within the range of variation of which its type falls (Jeffrey, 1968b). The correct name of a taxon would be the earliest name which was validly published since 1st May 1753 in any particular circumstance. Based on this rule therefore, we go back to check the correct names for the two wild species and the hybrid complex.

Cheesman (1948a, 1948b) gives us a very informative discussion on the possible earliest and correct names of bananas. He clarified that the genus *Musa* is typified by *Musa paradisiaca* L., a name published by Carl Linnaeus in his Species Plantarum (1753), the internationally accepted starting point for modern botanical nomenclature. Cheesman (1948a) considered this to be the first legitimate scientific name for the banana plant. *M. paradisiaca* was described as *Musa* with a nodding spadix. Linnaeus's protologue in the Species Plantarum 1: 1043 (1753) is as follows:

MUSA spadice nutante.....*paradisiaca.*

Cheesman (1948a) also stated that the above description was based on *Musa cliffortiana*, described in a tract *Musa cliffortiana* florens Hartecampi 1736 prope Harlemum. Cheesman (1948a, 1948b) continued that *Musa racemo simplicissimo* described in Hortus Cliffortianus (1737) was the same plant, about which Linnaeus added some further notes. The name *Musa paradisiaca* was typified by coloured plates of *Musa cliffortiana* published by Ehret in Trew's Plantae Selectae (1750). These plates were identified with a common cultivated type of plantain (AAB) with persistent bracts and male flowers by Cheesman (1948a, 1948b). *Musa paradisiaca* was therefore a cultivated seedless hybrid different from the wild diploids. The name *M. paradisiaca* then covers the hybrid complex which has been considered above.

In Systema Naturae (1759) Linnaeus provided a short diagnosis, to distinguish *M. paradisiaca* from a then imperfectly known banana he called *M. sapientum. M. paradisiaca* had persistent male flowers while the other banana did not. According to Cheesman (1948a) *M. sapientum* is typified by three plates showing the habit, the female flower and the ripe fruit of the same plant, drawn by Ehret and published by Trew (1750). Cheesman (1949) interpretes *M. sapientum* as also referring to an interspecific hybrid commonly known as silk fig (AAB). This is a second name published for what are now known to be AAB hybrids.

The two species *M. paradisiaca* and *M. sapientum* were considered two separate species by Linnaeus on the basis of having persistent male flowers. Cheesman (1948a)

argued that the persistence or non-persistence of male flowers on the rachis is not a sufficient character on which to separate species. He, however, believed that there were some other differences between *M. paradisiaca* and *M. sapientum* which warranted their being considered separate species.

A major concern about these two earliest names is that they were based on cultivated interspecific hybrids. The ICBN rules state that plants which have originated through interspecific hybridisation should have a scientific name which indicates this. For example *Musa paradisiaca*, the earliest published binomial, should be used for all products of crosses between the two wild diploid species. The epithet should be prefixed with a multiplication sign to indicate the hybrid nature of the species. *Musa sapientum* must be considered a synonym of the earlier *M.* x *paradisiaca*, since it applies to hybrids between the two wild species as well, but was published after *M. paradisiaca*.

A number of wild species and cultivars were discovered which did not fit the two Linnaean species. Subsequently many names were coined by scientists to describe the various forms. However, epithets are acceptable only if they conform to the ICBN. Furthermore, if a taxon is known by more than one name, it is the oldest validly published name which should be applied to the taxon under consideration.

One of the first names to be applied to a wild species was *Musa troglodytarum*. It was described in the second edition of Species Plantarum (1763) by Linnaeus as a species with an erect inflorescence. Pisang batu and *Musa uranoscopus*, species described earlier by Rumphius in 1750, were placed under *M. troglodytarum* by Linnaeus (Cheesman, 1948a). Linnaeus considered *Pisang batu* and *M. uranoscopus* as synonymous. Cheesman (1948a) disagreed with Linnaeus. *M. uranoscopus* had an erect inflorescence and *Pisang batu* had an inflorescence which was not erect and was likely to be the same species as the diploid wild species of bananas. Cheesman (1948a) rejected the name *M. troglodytarum* on the ground that it was applied to two different species.

Cheesman (1948a) stated that Loureiro in Flora Cochinchinensis (1790) described a number of species from Indo-China. The identity of some has never been certain because they were briefly characterised and could only be recognised by their native names. One of these was *M. seminifera* described by Loureiro as a wild form with seeds (Merrill, 1935). The same species was reported by Cheesman (1948a) to include another banana, Pisang Utan, described by Rumphius as a larger kind of the wild banana. This larger kind of the wild banana was associated with different forms of *M. textilis* (Anonymous, 1894). Loureiro cites elements now assigned to 2 different species. Pisang batu is now identified with *M. balbisiana*, Pisang utan is now identified with *M. textilis*. Loureiro's name is rejected as a nomen confusum (confused name). Cheesman (1948a) further mentioned that in the same Flora Cochinchinensis, Loureiro (1790) described *Musa corniculata* based on *Pisang Tando* of Rumphius. This species was quite distinct because it had fewer fruits which were very long and big. The description of the species did not apply to the two wild diploids under discussion. It was a horn plantain which is an AAB and therefore covered by Linnaeus's name *M. paradisiaca*.

Musa nana was another species described by Loureiro (1790) in the same Flora Cochinchinensis (Anonymous, 1894; Merril, 1935). According to Loureiro, this was a native of Cochin-China where it was called *Chuoi duii* (Anonymous, 1894). Baker (1893) thought it was the same species as *M. cavendishii* which was described by Lambert in 1837. However, Simmonds (1966) pointed out that although *M. nana* had been held by some authors to be the same as *M. cavendishii*, there were some features in the description which did not agree with this view. Baker (1893) described it as a form of

M. cavendishii with a taller stem and with fertile flowers. The feature of fertile flowers could be the difference Simmonds referred to. Further critical studies of Indo-China species were recommended by Cheesman (1948a) for clarification over Loureiro's species. The typification of the names of Loureiro's species is not certain.

Moore (1957) reported another species *M. rosacea* which was described by Jacquin in Plantarum Rariorum Horti Ceasarei Schoenbrunnensis (1804). Moore considered the name *M. rosacea* to have been misapplied to M. ornata Roxburgh since 1822. He stated that *Musa ornata* was listed in Roxburgh's Hortus Bengalensis (1814). *M. ornata* was however, described and published by William Carey in Flora Indica, 1824. Appended to this description was a footnote by Nathaniel Wallich that this was probably *M. rosacea* Jacq. which had been well figured in the Botanical Register (1823) 9: 706. Since then, the name *M. rosacea* has been misapplied by various workers. The plant which was described in Flora Indica was *M. ornata* but the name attached was *M. rosacea*. Cheesman (1948a) pointed out that true *M. rosacea* was an edible seedless banana which was closely related to one of the two wild species of bananas and he suggested that *M. rosacea* could be a hybrid between the two wild species of banana. *M. rosacea* was probably not one of the two wild species but a species to be covered by Linnaeus's earliest name, *M. paradisiaca*.

Cheesman (1948a) continued that between 1750 and 1820 nobody seems to have given the description of any wild banana like the one of Colla. Cheesman mentions Desvaux who described some wild species but Desvaux regarded all *Musa* species as varieties of *M. paradisiaca*. Cheesman (1948a) stated that Colla was the first to recognise *M. balbisiana* and *M. acuminata* as wild species distinct from any other species so far described. *M. balbisiana* was typified by Rumphius's species *"Pisang batu"* in the Herbarium Amboinense (1750). The description, figures and names were of pre-Linnaean time which Colla used to validly publish *M. balbisiana*. Likewise *M. acuminata* was typified by Rumphius's species *M. simiarum*. Colla's names are accepted as the valid names of the two wild species. *M. balbisiana* and *M. acuminata* were described in Memoria sul Genere *Musa* (1820, quoted by Cheesman, 1948a). Colla recognised *M. balbisiana* as different from those species previously described by Linnaeus which were edible species. However, Colla's type of *Musa acuminata* was one of the naturally occurring edible forms since it had no seeds. The following is Colla's brief description of the two species quoted from Cheesman (1948):
Musa balbisiana Colla, in Memorie della Reale Accademia delle Scienze di Torino 25:384 (1820) ("Memoria sul Genere Musa e Monografia del Medesimo").

M. spadice nutante, corde progerminante, floribus sterilibus et masculis deciduis, fructu semper virente polygono dorsis inaequalis. N.

Musa acuminata Colla, in Memorie della Reale Accademia delle Scienze di Torino 25:394 (1820) ("Memoria sul Genere *Musa* e Monografia del Medesimo")

M. spadice subnutante, floribus sterilibus et masculis deciduis, folis longe petiolatis undulatis, fructu sub-cylindraceo acuminato. N.

2.4.6 Infraspecific classification of bananas and plantains

The problems of infraspecific classification in both wild and cultivated plants have been discussed and debated quite widely for quite some time (Styles, 1986). Many authors reached a consensus that there are limitations to the formal hierarchy at the infraspecific level and this was largely because variation at this level is so complex, multidimensional and no longer hierarchical (Heywood, 1986). There were three major

problems which needed to be resolved particularly at the infraspecific level of cultivated plants and these were: whether wild plants and cultigens should be treated as separate species; whether special categories should be used for cultivated crops; and what approach should be considered below the subspecies level.

The International Code of Botanical Nomenclature (ICBN) recognizes five infraspecific ranks: subspecies, variety, subvariety, form and subform, although there is considerable debate over the most appropriate use of these and other infraspecific categories (Stace, 1980). The majority of plant taxonomists consider only subspecies, variety and form. Subspecies is used for geographical races, variety is used for local populations which are morphologically and possibly ecologically distinct and the form is used for atypical individuals within a population. The three categories may be used for cultivated plants as well as wild plants.

The genus *Musa* comprises both cultivated and the wild taxa. The cultivated taxa have been spread by human agency far from their original centres. They no longer show any natural range like the wild plants. The wild species have natural ranges. Some are widespread and overlap each other while others have restricted ranges (Simmonds, 1990a). In this circumstance, a subspecies which is understood in terms of geographical ranges cannot work for cultivated taxa since cultivated plants no longer have a natural distribution.

In bananas the rank of subspecies is used to distinguish the geographical races found within the wild *M. acuminata*. While this would agree with the conventional use of subspecies in wild plants, the subspecies later became something used to distinguish wild from cultivated taxa (Polhill & van der Maesen, 1985; Perrier & Tezenas du Montcel, 1988). The recent International Code of Nomenclature Cultivated Plants (ICNCP) (Trehane *et al.*, 1995) no long permits this. The categories of the ICBN should be used at or below the level of genus "if and only in so far as the crop species are identifiable with the botanical taxa in these ranks" (Trehane *et al.*, 1995). Instead, the ICNCP provides two alternative categories, cultivar group and cultivar. These are available only for cultivated plants and they are hierarchical. The nomenclature for cultivated plants can now be covered by the ICNCP.

Hanelt (1986) argued against using independent categories for cultivated plants. Hanelt argued that the conditions given by Jeffrey (1968a) for treating cultivated plants differently can apply to wild plants as well and are therefore no justification for treating them differently. He then discussed multiple classifications as the most useful principle of the infraspecific classifications. Lewis (1986) also pointed out that we should be encouraged to use the botanical hierarchy as far as that may be possible in cultivated plants.

The two categories which have been introduced by ICNCP, however, do not seem to cover the variation existing between cultivar groups and cultivars. This view is supported by a number of authors (Jeffrey, 1968a; Harlan & de Wet, 1971; Pickersgill, 1986b). Jeffrey (1968a) initially gave profound reasons for suggesting a different treatment of cultivated plants: they lack a true population structure, they do not occupy natural areas, they have a range of variability larger than wild plants, they suffer from a breakdown of isolation barriers thus favouring hybridization and introgression.

Other authors argued that two categories for cultivated plants are adequate (Brandenburg & Schneider, 1988) and that the categories start from cultivars as the basal culton (taxon of cultivated plants) so that assemblages of similar cultivars form groups. Cultivar groups may be based on one or more user criteria. One cultivar may belong to more than one cultivar group. For example in bananas Kayinja belongs to ABB genome group but,

being used for production of beer, may also be placed in a Beer group or Beverage group. They considered these as categories in an open classification i.e. recognition of one cultivar within a crop species does not mean that all other specimens within that species are automatically included in a second cultivar.

Pickersgill (1986b) considering the variation within a species argued that it is possible to have hierarchical categories of cultivated plants. Pickersgill (1986b) considered that there are three principal levels of change associated with domestication of crops, which can assist in creating hierarchical categories between species and cultivar. These are changes associated with domestication itself; subsequent partition of variation in the cultigen through geographical isolation, ecological adaptation, or human selection for different usages; and differentiation into the cultivars or landraces which constitute the actual field populations. Pickersgill (1986a) therefore suggested three categories which should treat these levels of variation: convarietas, group (subgroup if necessary) and finally cultivar or race.

It is indeed desirable to have more than two categories between a species and a cultivar for the reasons elaborated by various authors and they can be hierarchical (Jeffrey, 1968a; Harlan & De Wet, 1971; Parker, 1978; Pickersgill, 1986a). The question is how many of these are needed to cover the variability considered for any crop. These will vary depending on the variability existing in any given crop.

In many crops including bananas a number of informal categories have been introduced by various authors who have been working on the different crops to cover the variation between cultivar groups and cultivars.

The existing classification of bananas is that of Simmonds and others (Simmonds & Shepherd, 1955; Stover & Simmonds, 1987) based on genome groups below the level of the genus, a classification which has been widely and generally adopted for many years. The group as already indicated is a category acceptable under the International Code of Nomenclature for Cultivated Plants. Within these genome groups, many differences exist partly due to contributions of A genome from different wild subspecies of *M. acuminata* and partly due to mutations. The significance of mutations in bananas is very great because the number of clones has gradually increased in this way (Robinson, 1995). It therefore became apparent that all clones which have evolved through these mutations and through time needed some order. This variation is large and calls for some categories between cultivar group and cultivar.

The classification of *Musa* AAB into subgroups and clones was accomplished by De Langhe & Valmayor, 1980; Swennen & Vuylsteke, 1987; Swennen, 1988 and Lebot *et al.*, 1994. Example of subgroups are Plantains, Popoulu and Maia Maoli (Pacific plantains), Mysore, Silk, Pome and Pisanga Raja. The plantains were further divided into French (with persistent male bud) and Horn (with no male bud) (Simmonds, 1966), and the Horn subdivided further into three (Tezenas du Montcel *et al.*, 1983) as French Horn, False Horn and Horn types. Maia Maoli and Populu (Pacific plantains) were divided into clones and morphotypes (Lebot, 1994). The point here is, different workers recognise the same number of hierarchical categories but often use different names for these categories.

The classification of *Musa* AAA group into three subgroups Cavendish, Gros Michel and Green Red was proposed by Cheesman (1933). The Cavendish clones are thought to have derived from a single clone which diversified by somatic mutation (Daniells, 1990). Stover and Simmonds (1987) grouped the mutants into four: Dwarf Cavendish, Giant Cavendish, Grande Naine and Lacatan. These may probably be morphotypes according to Lebot (1994). The ranks of these are still not clear.

Shepherd (1957) proposed a fourth subgroup in *Musa* AAA group for the Lujugira-Mutika clones of the East African Highland region. He divided the Lujugira-Mutika subgroup into two. 'Mutika' consisted largely of clones with more or less pendulous, heavy bunches of medium to longish fat fruits which were conspicuously bottle necked, even at maturity. 'Lujugira' was characterised by subhorizontal bunches with shorter, less bottle-necked fruits. The two 'Mutika' and 'Lujugira' were never ranked. Sebasigari (1987) differentiated the beer and the cooking clones among the Lujugira-Mutika subgroup but never gave them ranks either.

The current classifications of bananas into and below genome groups are only as complete as the information available when they were constructed. For example, there are numerous cultivars and clones in Borneo and Indonesia which have not been classified (Robinson, 1996). All taxonomic treatments of bananas recognise Simmonds' genome groups. Some recognise the different subgroups but there are uncertainities about different taxa at the level of subgroups and below. Disagreements may arise from differences in taxonomic approach or differences in knowledge of the variability of different genome groups and subgroups. It is important to know to what extent a group varies and then determine and name units which can be used for reference and communication purposes.

The informal classifications used by banana workers have indicated that they employ at least up to three categories above the cultivar. In bananas the category group as discussed above is used to designate genomic groups like AAB. The subgroup is then used to differentiate for example the Plantains, the Silk and the Pome within the AAB. It seems that one more rank is needed to cover the variation that is found to exist in the different subgroups such as the Plantains or the Cavendishes. This will accomodate taxa such as French or Horn Plantains which are below the rank of subgroup but above the cultivar (Pickersgill, 1995b). There is no such category provided by the ICNCP. Besides a number of workers will differ in their opinions as to the number ranks that ought to be recognised while others will differ in naming the various categories. There will always be lack of uniformity in ranking and naming of these categories since variation of each crop is different.

In view of the problems connected with the infraspecific classification of bananas and other crops, a phenetic approach will be adopted in the study of the East African Highland bananas to survey the variability in the subgroup and to determine units for purposes of reference and communication. The units will then be named.

The Highland bananas are clones and it has been accepted that they have diversified through mutations and distinct mutations are therefore clones (Trehane *et al.*, 1995). Farmers' information, and researchers' knowledge of the crop suggest that there are possible groupings among the Highland clones. Groupings among the Highland banana clones are necessary for ease of communication and to assist choice of diverse clones for assessment of disease and pest resistance. The Highland bananas have been recognised and referred to as a subgroup (Shepherd, 1957; De Langhe, 1986). Clones of the East African Highland bananas can be grouped into some higher order category above clone but below subgroup. This category will be called clone set.

Clone sets are expected to be useful provided that other facts can be integrated into them as information becomes available. Categories like these clone sets, to be achieved through the use of phenetic analyses, do result in a more natural grouping of the infraspecfic variability (Baum, 1981; Hanelt, 1986). They have a predictive element in them, a factor which has always been advantageous in most natural classifications.

Chapter 3

ACCESSIONS, CHARACTERS AND DATA ANALYSIS

3.1 Introduction

This part of the work covers the general methods which were carried out in the different phases of the study. They include selection of accessions, selection and scoring of characters and analysis of the data. The clones included in the study were growing in the national banana germplasm collections. A brief history and information on the management of these collections is given for purposes of later discussions.

3.2 National banana collections

3.2.1 Historical review

In Uganda, the first banana germplasm collection was established in the 1920s at Kampala plantation by Maitland (Tothill, 1940). It contained cooking, roasting, beer and dessert types of bananas collected from the central and western parts of Uganda. The collection was transferred to Kawanda Agricultural Research Institute in 1940 as land was getting scarce in the Kampala area. The number of accessions continued to increase so that it became necessary to transfer the collection further to Bukalasa experimental site, 25 kilometers to the north of Kawanda (figure 3.1).

The number of accessions had risen to 227 after more clones had been collected from all banana growing regions of Uganda (Western, Central and Eastern parts). The aims of collecting and maintaining a banana collection before the 1970s were to study synonyms of local names and to attempt to match the Ugandan bananas with those grown in other countries. A comparison with descriptions and photographs showed that many Ugandan bananas were also grown in Tanzania (Tothill, 1940). No further work was done on the collection between 1979 and 1986 due to political instability which led to the deterioration of the collection.

In 1989 two collections were re-established, by re-collecting, one at Kawanda and another at Kabanyolo. Since 1989, the two collections have been steadily increasing in size, while funds for maintenance have become steadily more difficult to find. Reduction in size of the collections is necessary for their efficient utilisation.

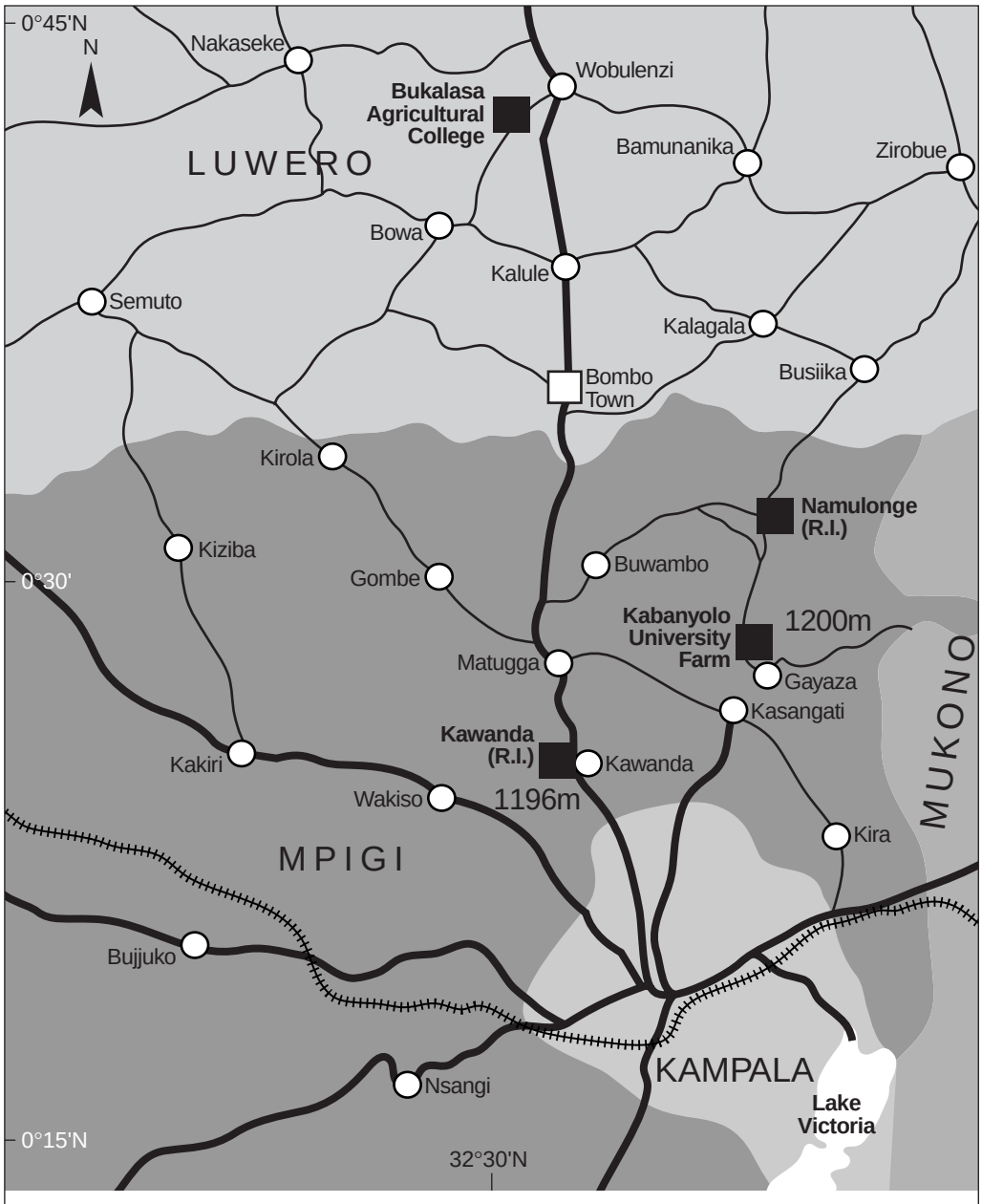


Figure 3.1: Locations of Bukalasa, Kawanda and Kabanyolo Agricultural Research Institutes.

3.2.2 Location and climatic background

The two collections are located in Mpigi district but twenty kilometers apart, Kawanda at 32° 32'E and 0° 25'N and Kabanyolo at 32° 37'E, 0° 28'N (Fig. 3.1). They are 1196-1200 metres above sea level. The climate is subhumid, with mean annual temperatures between 20° and 25°C. The mean annual rainfall is between 1120 and 1140 millimitres, bimodally distributed. The main wet season is March to May with short rains in September to November. The rainy seasons are punctuated by two dry seasons, one in January to February and the other in June to July. The soils are reddish-brown loams with pH between 5.1 and 5.8. The two collections are therefore basically in the same environment.

3.2.3 Management

Kawanda collection is managed by the National Agricultural Research Organisation and is used to study variation, provide material for breeding, and evaluate pest and disease resistance. Kabanyolo collection is under the mandate of Makerere University and is used mainly for training purposes.

Both collections were established with suckers which were planted in holes 30-40cm deep. In each collection an accession is represented by at least 10 plants planted in a single row, with a spacing of 3 x 3 metres within and between rows.

Although the collections are in a similar environment, they are managed differently. At planting SSP (Single-super phosphate) is applied in planting holes at Kawanda and cow dung at Kabanyolo. Otherwise chemical fertilizers are rarely used in either collection. At Kawanda pesticides against banana weevils and nematodes are applied regularly twice before the rainy seasons whereas at Kabanyolo pesticides are not applied routinely. At Kawanda fertility is maintained by mulching once a year at the beginning of the dry season with old banana leaves and grass (mainly *Pennisetum purpureum* L.), in addition to coffee husks, to a depth of 10cm. Mulching is rarely done at Kabanyolo and if it is done, coffee husks are applied, when they are available, unsystematically between rows. Deleafing and pruning have been practised regularly at Kawanda leaving 3 shoots per mat such that the 3 shoots grow and flower at 3 month intervals. This is not systematically done at Kabanyolo. By the time this study took place, the first three blocks of Kabanyolo collection had been eroded while the other blocks had some mulched and non mulched patches.

3.2.4 Accessions studied

The collections hold more than 400 accessions of different genome groups but predominantly East African Highland bananas. Accessions in the collections are classified into genome groups and the local name used for each accession is also recorded. The accessions are in different phases of ratoon cycles since they were all collected and planted at different times. Those which were in their third year ratoon cycle were selected for study, because they are then considered stabilised and will show the true characters of the clone (De Langhe, 1961). The analysis was also restricted to accessions which could provide 5 randomly selected healthy plants per accession.

204 accessions were thus selected for study from the national collections (Table 3.1). A preliminary analysis was conducted on sample of accessions from different genome groups, including the Highland bananas, grown at Kawanda. This preliminary analysis

Table 3.1: Accessions used in the analysis.
(C=Kawanda accessions; K=Kabanyolo accessions; F=Farmers' accessions; B=Beer accessions; M=Mutants; *=Ramets): c=from central region; w=from western region; e=from eastern region.

EAST AFRICAN HIGHLAND (AAA-EA)	
Code	**Local name**
Beer clone set	
CM4	Mudwale Beer(e)
KB49	Nakayonga(c)
*CB53, KB53	Namadhi(e)
CB54	Mende(c)
CB55	Bagandeseza(c)
KB56	Enyarukira(w)
*CB57, KB57	Nalukira(c)
*CB58, KB58	Endirira(w)
*CB59, KB59	Katalibwambuzi(w)
KB84	Oruhuuna Beer(w)
KB87	Namunyere Beer(e)
KB123	Enkara(w)
KB124	Enshenyuka(w)
KB125	Entanga(w)
KB126	Imbululu(e)
KB127	Bwara(w)
KB128, FB128	Enywamaizi(w)
KB129	Nametsi(e)
KB130	Nalusi(e)
KB131	Liyase(e)
KB132	Shombo-Obureku(w)
KB133	Kibagampera(e)
KB134, FB134	Engumba(c)
KB135	Enjumba(w)
KB136	Engambani(w)
KB137	Kaitaluganda(e)
KB138	Nalwesanya(c)
KB139	Endembezi(w)
KB140	Mutant mixed(w)
KB158	Mwanga(w)
KB159	Naminwe(e)
KB176	Ensika(w)
FB177	Engote(w)
FB178	Entundu(w)
FB179	Baleka(w)
FB180	Omuzo(w)
FB181	Nakanyala(w)
Musakala clone set	
C1, K1	Muvubo(c)
C2, K2, F2	Musakala(c)
C3	Nakibizzi(c)
C4, K4, F4	Mudwale Cooking(e)
C5	Luwaata(c)
C6, K6	Mayovu(e)
C7, F7	Kisansa(c)
*C8, K8	Mukazialanda(w)
C9, K9	Nalugolima(c)
C10	Muturit(e)
C1 1	Lumenyamagaali(c)
C12	Mugisuagenda(e)
*C13, K13	Wansimirahi(w)
C14, K14	Namunwe(c)
C15, K15	Siira Red(c)
KM15	Siira White(c)
C16, K16	Atwalira(c)
*C17, K17	Kabucuragye(w)
K90	Nassaba(e)
K141	Mpologoma(c)
K142	Rwamigongo(w)
K143, F143	Lwewunzika(c)
K144	Mujuba(w)
K145	Bandangeya(w)
K146	Rwabakongo(w)
F163	Batule(e)
F169	Enyoya(w)
F174	Namunget(e)
Nakabululu clone set	
C47,K47	Nakabululu(c)
F47	
*C48,K48	Nakasabira(e)
C49	Nakayonga(c)
C50,K50	Kazirakwe(w)
C51,K51	Butobe(w)
C52,K52	Kaitabunyonyi(e)
K115,F115	Bukumo(w)
K116	Enkonera(w)
K117	Kisugunu(e)
K118	Bifusi(e)
K119	Embururu(w)
K120	Kafunze(c)
K121	Wekhanga(e)
K122	Mukite(e)
F165	Mukubakkonde(c)
F175	Mburiondet(e)
Nakitembe clone set	
C18, K18	Namaliga(c)
C19, K19	Nakitembe(c)
C20, K20	Nakitembe-Nakamali(c)
*C21, K21	Namulondo(c)
C22, K22	Nakitembe-Nakawere(c)
C23	Nakitembe-Omunyoro(e)
C24	Nakitembe-Omumyufu(c)
C25	Nakibuule(c)
C26, K26	Mbwazirume(c)
C27	Nakyetengu(c)
CM27	Nakyetengu-Omuwanvu
K28	Ekitetengwa(w)

Table 3.1: Cont'd.

EAST AFRICAN HIGHLAND (AAA-EA)	
Code	**Local name**
C29	Kibuzi(c)
K72	Nandigobe(c)
K73, F73	Entaragaza(w)
K74, F74	Enjagata(w)
K75	Toro(w)
K76	Ntinti(w)
K77	Luvuta(e)
K78	Nalwera(e)
K79	Nabuyobyo(e)
K80	Nasaala(e)
K81	Nakangu(e)
K82	Enshakara(w)
K83	Rwakashita(w)
K84	Oruhuuna-Cooking(w)
K147	Lisindaalo(e)
K148	Enyarutere(w)
K149	Waikova(e)
K150	Bikowekowe(e)
K151, F151	Salalugazi(c)
F161	Mukite(e)
F170	Kitika(w)
F171	Kongowet(e)
F172	Endyabawali(w)
F182	Nakitembut(e)
Nfuuka clone set	
C30,K30	Nakinyika(c)
F30	
KM30	Enyakinika(w)
C31,K31	Nfuuka(c)
F31	
C32,K32	Namwezi(c)
F32	
CM32	Namwezi black(c)
K33	Kulwoni(e)
C34,K34	Ndyabalangira(c)
C35	Tuula-twogere(c)
C36,K36	Nakabinyi(c)
*C37,K37	Enyeru(c)
F37	
*C38,K38	Nakhaki(e)
C39	Tereza(c)
C40	Nakawere(c)
C41	Enjeriandet(e)
*C42,K42	Nante(e)
*C43,K43	Bitambi(c)
*C44,K44	Enzirabahima(w)
*C45, K45	Entukura(w)
C46, F46	Namande(c)
K85	Entazinduka(w)
K86	Enjogabakazi(w)
K87	Namunyere Cooking(e)
K88	Kasitaza(c)
K89	Kisaabo(c)
K91	Rwezinga(w)
K92	Mutta-Ngendo(c)
K93	Nabusa(c)
K94	Enkobe(w)
K95	Likhago(e)
K96	Kufuba(e)
K97	Rwasha(w)
K98	Sitakange(c)
K99	Serunjogi(c)
K100, F100	Namafura(e)
K101	Nasuuna(c)
K102	Kabende(e)
K103	Nyamanshari(w)
K104, F104	Nambokho(e)
K105	Nakijumbi(w)
K106	Mukaddealikisa(c)
K107, F107	Lusumba(c)
K108	Namamuka(e)
K109	Nambi(c)
K110, F110	Enyabakazi(w)
K111	Enyakagongo(w)
K112, F112	Khabusi(e)
K113	Nyabungere(w)
K114	Namakhumbu(é)
K152	Nzirabushera(w)
K153	Kasenene(w)
K154	Karinga(w)
K155	Kaasa(w)
K156, F156	Kibalawo(c)
K157	Enyaruyonga(w)
F160	Katwalo(c)
F162	Ntika(c)
F164	Nakibira(c)
K166	Nabusolo(e)
F167	Enshenyi(w)
K168	Enyambo(w)
F173	Rwambarara(w)
DESSERT AAA	
D60	Bogoya-Omweru
D61	Bogoya-Omumyufu
D62	Dwarf Cavendish
D63	Semi-Dwarf Cavendish
D64	Tall Cavendish
PLANTAIN AAB	
P65	Nakatansese
P66	Gonja Kakira
P67	Gonja Manjaya
BLUGGOES ABB	
S68	Kayinja
S69	Kivuvu
NEY POOVAN AB	
T70	Kisubi
T71	Sukali-Ndiizi

was in order to assess whether the classification produced by multivariate analysis of the characters chosen was similar to the widely accepted conventional classification of bananas into genome groups. Accessions used in this preliminary analysis included 59 East African Highland banana clones (AAA-EA), 5 dessert clones (AAA), 3 Plantains (AAB), 2 Bluggoes (ABB) and 2 hybrid edible diploids (AB). The prefix D was included in the code for dessert bananas (AAA), P for plantains (AAB), S for Bluggoes (ABB) and T for diploid hybrids (AB).

The phases of studies carried out were; 1) preliminary analysis including all genome groups available; 2) analysis of *Musa* AAA-EA in the Kawanda collection and 3) analysis *Musa* AAA-EA in the Kawanda and Kabanyolo collections. In the last analysis, 192 accessions of both cooking and beer bananas were used. The letter B is included in the identifying code for accessions of beer bananas. Clones of Highland bananas with the same local name were assigned the same number in the analyses, prefixed by C for accessions grown at Kawanda and K for accessions grown at Kabanyolo.

According to accession data, ramets from single individuals have been exchanged between the two collections. Accessions known to be ramets are marked with asterisks in Table 3.1. Some other accessions have similar names in the two collections but were collected independently from different sites.

Some somatic mutants have occurred since the accessions were collected. These would be taken away from the parent clone and propagated separately in a new row in the collection. These included CM27 (parent C27), CM32 (parent C32) from Kawanda collection, KM15 (parent K15) from Kabanyolo collection and hence were considered known mutants. The farmers claimed that some of their clones had arisen as somatic mutants. These included CM4 (Mudwale beer) and its presumed parent C4 (Mudwale cooking), KM30 (Enyakinika) mutant of K30 (Nakinyika). Some were collected as 2 separate accessions but with local names that indicate their presumed relationship but not necessarily mentioned by farmers that they were mutants. These included KB84 (Oruhuuna beer) and K84 (Oruhuuna cooking), KB87 (Namunyere beer) and K87 (Namunyere cooking). Known somatic mutants were designated with an additional letter M in their identifying code. All the Highland bananas were assigned to clone sets in a subjective classification by the researcher, based on few selected quickly observable characters. The subjective clone sets with characters used are shown in Table 3.2. Clusters generated from the multivariate analyses were then compared with this subjective classification.

In a final study, 46 accessions from farmers' fields in selected sites were added to the analysis. These consisted of 23 accessions which shared names with one or more accessions from the two collections and 23 accessions with local names not represented in the collections. All accessions studied in farmers' fields were prefixed by F. The farmers' fields were located in three regions: the eastern highlands, the south western highlands (both at 1400-1800 meters above sea level) and the central lowlands (below 1400m). The major differences between the three regions which would directly affect plants under study are the altitude, hence the cool temperatures prevailing in the highlands, and management of the crop. Most bananas in the south western regions were mulched with grass and old banana leaves whereas the majority of those in the eastern highlands and central lowlands were not. Desuckering was systematically practised in the south western region but not in the eastern or central regions. Weeding by hoe was common in the lowlands and eastern highlands but farmers in the south western region would remove the weeds by hand. Most farmers were not using pesticides but different cultural methods were employed to control weevils and nematodes.

Table 3.2: The subjective classification of the East African Highland banana clones.

Clone sets	Description
Beer	Pulp colour before maturity white with sticky brown excretions across the pulp, pulp colour after maturity cream with sticky brown excretions across the pulp. Unripe pulp bitter and astringet. The clones can have characters of other clone sets but the pulp differs.
Musakala	Pulp colour before maturity white with no sticky brown excretions and cream with no brown excretions after maturity. Unripe pulp insipid. Pendulous, lax bunches, long fruits above 20cm. Fruits with bottle necked apices, pendulous nude male inflorescence rachis.
Nakabululu	Pulp colour before maturity white with no brown excretions and creamy brown with no brown excretions after maturity. Unripe pulp insipid. Subhorizontal to very compact bunches, short fruits below 15cm with almost blunt apices. Subhorizontal to oblique nude male inflorescence rachis.
Nakitembe	Pulp colour before maturity white with no brown excretions and creamy brown with no brown excretions after maturity. Unripe pulp insipid. Oblique compact bunches, medium fruits 15-20cm long. Fruit apices intermediate (between bottle necked and blunt). Oblique male inflorescence rachis with persistent bracts and neuter flowers.
Nfuuka	Pulp colour before maturity white with no brown sticky excretions and cream with no brown excretions across the pulp after maturity. Unripe pulp insipid. Oblique compact bunches, medium fruits 15-20cm long. Fruits with apices intermediate (between bottle necked and blunt), oblique or sigmoid curved nude male inflorescence rachis.

3.3 Character selection and scoring

A character is a feature of an organism that can be measured, counted or otherwise assessed (Heywood, 1967). Characters form the central theme of any study concerned with identification and classification of organisms. A good classification of objects will largely depend on the characters selected, how varied they are, whether they show discontinuities and also the way they are treated (Pankhurst, 1991). Characters may not always be of equal value for purposes of comparison for example characters which do not vary in a group under study are not useful because they are unable to separate them, while characters which are logically correlated are neither acceptable because they provide similar information i.e different ways of expressing the same thing (Pankhurst, 1991). Characters are chosen on criteria such as ease of observation, availability and usefulness in classifying and identifying organisms. Morphological characters are commonly used in identifying plants.

Good characters are not easily modified by environmental factors and have a genetic basis such that they are unlikely to change readily (Heywood, 1967; Jeffrey, 1968b). These may be referred to as constant characters and are highly heritable. On the other hand, bad characters are easily modified, to a greater or lesser extent, by the environment. Their phenotypic expression is the product of the combined effect of the environment and the genotype. They are not constant and hence not dependable when classifying organisms.

Selection of characters to use in grouping organisms is a matter which has given rise to much dispute. In phenetic classification, it has been argued that as many characters as possible ought to be used when classifying organisms (Sneath & Sokal, 1973; Pankhurst, 1991). This will give a classification based on the overall similarity of organisms. Phenetic classifications usually reflect at least partially the phylogeny of the group under study since overall similarities are often due to descent from a common ancestor, except in cases of convergence or parallelism.

In phylogenetic or cladistic classifications it is argued that only characters which bear evolutionary significance must be used, and other characters must be ignored (Pankhurst, 1991). This then raises the problem of how to select such characters. Such classifications will take a long time to achieve because we cannot get any direct evidence of the whole course of evolution of any living organism as the fossil record is very incomplete. However, cladistic classification can make various assumptions about the direction of evolution in any given character, they do not wait to discover fossil records. Molecular systematists accept that DNA can provide evidence of the course of evolution in any group but molecular data are still far from complete.

Pheneticists believe therefore that a phenetic classification would be appropriate and in order to produce such classification we should start with as many characters as possible giving equal weight to each character. Characters should come from every part of the plant not just a sample from particular parts of the plant presumably because different parts of the genome affect different suites of characters and at different times. Another reason for using a large number of characters is because only a few characters are likely to be causally correlated or prone to convergent evolution.

Complex characters are broken down into unit characters with the aim that each unit character shall as far as possible contribute one new item of information that is relevant in building up a phenetic classification. Sneath and Sokal (1973) defined a "unit character" as a "taxonomic character of two or more states which cannot be further subdivided logically except for subdivision brought by changes in the method of coding the states."

With this background therefore, characters to be used in this study were selected based on ease of observation, availability, and discontinuity. Complex characters were broken down to unit characters.

3.3.1 Characters scored and methods of scoring

73 characters were selected to be used in the study (Table 3.3). Characters included those used by Simmonds (1959, 1962, 1966), Purseglove (1972) and Stover and Simmonds (1987) to distinguish clones which contain a *balbisiana* genome from those which are of *acuminata* origin only. Other characters were those used by Shepherd (1957), Sebasigari (1990), Rossel & Mbwana (1991) and IBPGR (1984) and IPGRI (1996) descriptors to distinguish the Highland bananas from other groups (AB, AAB, AAA and ABB). Still others were based on the researcher's experience with the variation found in the East African Highland bananas and those characters used by farmers.

Characters included sixty qualitative and thirteen quantitative characters. Twenty characters were derived from vegetative parts, twenty eight from the female inflorescence (bunch and fruits) and twenty five from the male inflorescence (Table 3.4).

3.3.1.1 Coding

Simple two-state qualitative characters e.g watery sap or milky sap, were coded as binary e.g by scoring one for watery sap and two for milky sap. Ordered multistate characters were coded as series of discrete states. For example if there were three flower colours, red, pink and white, individuals could be scored one for white, 2 for pink (intermediate between red and white) and 3 for red. Low intensities of a colour or any other character were given lower scores so that scores would increase with increasing intensity of a colour or any other character.

Table 3.3: Characters used in numerical analysis.

*** characters useful in distinguishing clones containing *balbisiana* genome.**
**** characters useful in distinguishing the East African Highland bananas).**

No.	Characters (with their coded states if qualitative)
Vegetative structures	
1.	Pseudostem height/girth ratio.
2.	Pseudostem background appearance (1.bright coloured 2.dark coloured).
3.**	Pseudostem (1.not glossy 2.glossy).
4.	Pseudostem undersheath colour (1.yellow-green 2.greyed-yellow 3.not applicable).
5.	Pseudostem undersheath anthocyanin (1.absent 2.present).
6.**	Pseudostem blotches (1.absent 2.present).
7.**	Blotch colour (1.brown 2.bronze 3.black 4.not applicable).
8.	Sap colour (1.watery 2.milky).
9.	Sap dripping (1.does not drip 2.drips).
10.	Suckers with tubular\scale leaves at 1.5-2.0 m high (1.non-tubular 2.tubular).
11.	Sucker orientation (1.not growing at an angle 2.growing at an angle).
12.*	Petiole margins (1.inrolled 2.erect 3.spreading).
13.	Petiole background colour (1.watery green 2.green 3.not applicable).
14.	Petiole anthocyanin (1.absent 2.confined to margins 3.throughout ventral side).
15.	Petiole length/width ratio.
16.	Distance between petiole bases.
17.	Leaf length/width ratio.
18.	Leaf tip (1.not twisted 2.twisted).
19.**	Leaf colour (1.yellow green 2.dirty green 3.glossy green 4.not applicable).
20.	Leaf anthocyanin (1.absent 2.present).
Female inflorescence structures	
21.	Peduncle length/girth ratio at harvest.
22.*	Peduncle hairiness (1.glabrous 2.finely hairy 3.coarsely hairy).
23.	Bunch orientation (1.sub-horizontal 2.oblique 3.pendulous).
24.	Bunch length/circumference ratio at harvest.
25.	Bunch shape at harvest (1.rounded 2.rectangular 3.cylindrical 4.truncated).
26.	Bunch compactness (1.lax (hand internode greater than 10 cm) 2.compact (hand internode 5-10 cm) 3.very compact (hand internode 5 cm or less).
27.	Fruits arrangement (1.uniseriate 2.biseriate).
28.	Fruit fusion (1.not fused 2.fused).
29.	Fruit/hand ratio (no. of fruits of 2nd hand $\prod$ numbers of hands in a bunch).
30.	Fruit position in the bunch (1. positively geotropic (strongly recurved to touch rachis) 2.positively geotropic (less strongly recurved towards but not touching rachis) 3. no geotropic reaction (fruits perpendicular to rachis) 4.not applicable).
31.	Hand arrangement (1.no internodes, continuous spiral of fruits 2.internodes present

Table 3.3 Cont'd.

No.	Characters (with their coded states if qualitative)
	between successive hands of fruits).
32.*	Fruit skin (1.non waxy 2.waxy).
33.	Fruit skin colour (1.variegated (green and yellow) 2.green 3.glossy green 4. not applicable).
34.	Fruit anthocyanin (1.absent 2.present).
35.	Fruit width/pericarp thickness ratio.
36.	Ovules (1.absent 2.present).
37.*	Ovule rows in the loculus (1. 2 rowed 2. 4 rowed 3.not applicable).
38.	Pulp colour before maturity (one month after shooting) (1.white 2.cream 3.orange brown).
39.	Pulp colour at maturity (4 months after shooting)(1.white 2.cream 3.orange brown).
40.	Pulp (1.without brown sticky excretions 2.with brown sticky excretions).
41.	Pulp taste (1.insipid 2.astringent and bitter).
42.	Fruit apex (1.blunt 2. blunt to bottle-necked 3. bottle-necked).
43.	Style on fruit apex (1.non-persistent 2.persistent).
44.	Type of persistent style on fruit apex (1.dry style 2.fleshy style 3.not applicable).
45.	Stamens (1.non-persistent 2.persistent).
46.	Fruit cracking (1.no 2.yes).
47.	Fruit length/width ratio.
48.	Fruit shape (1.rounded 2.rectangular 3.triangular 4.gourd-shaped 5.slender).
Male inflorescence structures	
49.	Male bud (1.absent 2.present).
50.	Male inflorescence rachis position from bunch (1.sub-horizontal 2.oblique 3.pendulous 4.not applicable).
51.	Male inflorescence rachis anthocyanin (1.absent 2.present 3.not applicable).
52.	Male inflorescence rachis length.
53.	Male neuter flowers along the rachis at harvest (1.non-persistent 2.semi-persistent 3.persistent 4.not applicable).
54.	Male inflorescence rachis nodes.
55.	Male bud anthocyanin (1.absent 2.present 3.not applicable).
56.**	Male bud colour (1.crimson 2.purplish-blue 3.bluish-purple 4.not applicable).
57.	Male bud shape (1.elliptical 2.lanceolate 3.oblong 4.cordate 5.ovate 6.not applicable).
58.	Male bud waxiness (1.non-waxy 2.intermediate 3.waxy 4.not applicable).
59.	Male bud apex (1.pointed 2.intermediate 3.obtuse 4.not applicable).
60.	Bract imbrication (1.not imbricate 2.imbricate 3.not applicable).
61.	Bract length/width ratio.
62.	Bract curling (1.not rolled back 2.rolled back after opening 3.not applicable).
63.*	Colour of internal face of bract (1.fading to yellow towards the 2.crimson towards the base 3.not applicable).
64.	Length of male flower/length ovary ratio.
65.	Compound tepal (1.not tubular 2.tubular 3.not applicable).
66.*	Lobes of compound tepal (1.white 2.yellow 3.orange 4.pink 5.not applicable).
67.*	Basal parts of compound tepal (1.not pigmented 2.pigmented 3.not applicable).
68.	Colour of compound tepal (1.yellow 2.pink 3.not applicable).
69.	Free tepal shape (1.oval 2.rectangular 3.fan shaped 4.not applicable).
70.	Free tepal at basal margins (1.not serrated 2.serrated 3.not applicable).
71.**	Pollen sac colour (1.white 2.yellow 3.orange 4.pink 5.not applicable).
72.	Filament hooked (1.not hooked 2.hooked 3.not applicable).
73.	Stigma colour (1.white 2.cream 3.yellow 4.orange 5.not applicable).

Table 3.4: Type of characters used in the analysis.

Type of characters	Vegetative structures	Bunch & fruit	Male bud	Total
Quantitative	4	5	4	13
Qualitative	16	23	21	60
Binary	10	13	11	34
Multistate ordered	6	8	8	22
Multistate unordered	0	2	2	4
Distinguish genome group	1	3	7	11
Distinguish East African Highland clones	3	0	2	5
Total	**20**	**28**	**25**	**73**

A few non-ordered characters were coded as series of discrete states because ratios were not giving a true picture of their shapes. These were bunch shape, fruit shape, male bud shape and free tepal shape.

Characters related to colour were mainly examined indoors. The standard Royal Horticultural Society (1986) colour chart was used in colour scoring. Cut pieces of leaves, pseudostems, tepal lobes, stamens, stigmas, fruit skins, pulp

and male buds were examined under the hole in the colour patch of the RHS chart so that natural colours could easily be matched with the colour of the chart. The colour numbers of the chart displayed in appendix 1 are the standard expression of the described colour state of that character.

If there was variation within an accession for qualitative characters, the overall score considered was for the majority of the five plants under study i.e three or four out of five plants scored.

Quantitative characters were entered directly as raw data. They were measured to the nearest centimetre or nearest millimetre using calibrated tapes. Mean values for continuous quantitative characters for the five randomly selected healthy plants per accession were calculated and most quantitative data were converted to ratios to reduce environmental effects.

3.3.1.2 Missing data and inapplicable characters

Missing data were coded as 999 as recommended by Rohlf (1993). For example the five plants which lacked male buds because farmers had cut them, had missing data. However, some accessions had no male buds due to genetic reasons. This was a character in its own right and was scored as a binary character: male bud absent or present.

Other cases where characters were unable to be scored, involved dependent characters. For example in accessions with no male buds, “shape of male bud” could not be recorded. Shape of male bud is a dependent character, inapplicable for accessions which never had male buds. Other characters could not be scored because they were masked by others. For example in accessions which have red petioles and midribs, the shade of green present is masked by the red colour and cannot be scored. Characters of these types were coded 999, indicating inapplicable.

3.3.2. Vegetative characters

The vegetative characters were scored within fifteen days of inflorescence emergence. During this time, maximum development of vegetative parts takes place (Purseglove, 1972; Karamura and Karamura, 1995).

3.3.2.1 Pseudostem and sap

The pseudostem is the false stem of the banana plant, consisting of enclasping leaf sheaths. The pseudostem varies in height, girth and colour. Pseudostem height was measured from the level of root emergence to the level of inflorescence emergence from the pseudostem. This is the point where the peduncle (bunch stalk) comes out of the pseudostem before it bends to support the bunch. Pseudostem girth was measured at 20 cm from the level of root emergence and the ratio of plant height to girth was used.

Background colour of the pseudostem varies in different genome groups. Clones of AB, AAB, ABB and AAA dessert bananas have bright green pseudostems while most East African clones have extensive mottling on their pseudostems almost masking the green colour of the stem. The background colour was assessed using a colour chart from a distance of four meters away from the plant.

The leaf sheaths of bananas clasp around the aerial stem which carries the inflorescence. The outermost sheaths are loosely clasping and can be removed. Two outer sheaths were removed to expose the inner sheath. At the base of the inner sheath, the colour was homogeneous and constant. Most clones of the AAA group have anthocyanin in this part of the plant. Presence or absence of anthocyanin was recorded at a height of 20 cm above soil level.

The East African Highland bananas are characterised by intense purple/brown /bronze /black mottling of the leaf sheaths (also called blotches). A few clones are not mottled at all while others have varying degrees of mottling. The colour of the blotches also varies although black predominates.

Among the Highland bananas are clones whose sap does not drip when the pseudostem is wounded or when a leaf is cut from the plant, while others start dripping almost immediately they are cut. The colour and viscosity of the dripping sap varies from near-clear watery to milky viscous liquid.

3.3.2.2 Banana suckers

Banana suckers are lateral shoots from the main banana plant which can develop into a bearing plant. There is variation in the degree of production of suckers, in types of leaves produced and position of emergence from the soil.

The last two characters were scored. The first character needs a study of several cycles of the plant. Banana suckers normally produce narrow scale leaves until they are one metre high. If the suckers are damaged, for example through weevil attack, the scale leaves quickly turn into normal leaves (water suckers). However, in some Highland banana clones, suckers consistently continue to produce scale or tubular leaves (Rossel & Mbwana, 1991) even up to 2 meters above the ground. Only one accession was found to have this character.

Two accessions were found to produce suckers which emerge at an angle to the mother pseudostem whereas in most clones the suckers emerge from the soil in an erect position parallel to the mother stem.

3.3.2.3 Petiole

The petiole is the stalk of the leaf between the sheath and the blade. It varies in length, width at its base, margin colour, whether the margins are spreading, erect or inrolled, background colour and presence of anthocyanin.

The length of the petiole was taken by slotting an adjustable ruler into the petiole groove of the 3rd or 4th leaf below the bunch (Shepherd, 1957). It was measured from the adaxial side, starting from the point of attachment on the stem to the base of the lamina. The width was recorded from the abaxial side, at the point of detachment of the petiole from the stem round the curved base (widest point) to the opposite side of detachment from the stem.

Farmers also attempt to differentiate some clones of the East African Highland bananas by assessing distances between petiole bases. Distance between petiole bases was scored from the lowest petiole of the lowest fresh leaf for 3 successive leaves on each of the five plants per accession, at the point where the petiole is detaching itself from the stem to the next petiole base above the first one. Two distances were taken on each of the five plants to get the average. The 2 lowest distances are always constant after shooting.

3.3.2.4 Leaf blade

Leaves vary in length, width, form of leaf tip, colour and anthocyanin presence. Leaf length and width were measured after cutting the leaf. The measurements were taken on level ground immediately after cutting to avoid shrinking. The fourth leaf below the bunch was measured (Shepherd, 1957). The leaf length was scored from the lamina base to the lamina tip. Width was scored at the widest part of the leaf.

In some clones the cigar or funnel leaf (the youngest central leaf of a banana plant which is still unfolded) becomes twisted at the tip and at maturity the leaf tip gets a wavy or twisted appearance. This character is common in a subset of the Highland bananas.

Leaves of the banana crop have various shades of green, but there are some which are red or purplish in colour indicating anthocyanin presence. Anthocyanin presence was best scored during the rainy season because dry weather tends to reduce pigmentation.

3.3.3 Bunch and fruit characters

Bunch and fruit characters were scored three months after shooting when a bunch showed a ripened finger (a single individual banana fruit) on the first hand (a cluster of fingers). At this stage the bunch is mature and ready for harvest.

Bunch and fruit characters vary greatly because of selection pressures which affect them more than any other parts of the plant. Sizes of fruits are of commercial importance although flavour of different clones does not necessarily correspond with size of fruits. Buyers who know clones with a particular flavour they prefer, are the only ones who may pick what exactly they want. Otherwise the majority of buyers chose bunches with large fruits. Characters which vary include peduncles, orientation, length, circumference, shape and compactness of the bunch.

3.3.3.1 Peduncles

Peduncles (bunch stalk) vary between clones in length, girth and hairiness. The length was recorded from the point of emergence from the pseudostem to the node of the first hand in the bunch.

The girth was taken at the point of emergence of the peduncle from the stem. Peduncle hairiness is a character important in discriminating clones which contain an *acuminata* genome (Simmonds, 1966). Groups ABB and AB have a glabrous peduncle, those of AAB are finely-hairy and the AAA triploids, especially the Highland bananas, have coarsely-hairy peduncles. The degree of hairiness was scored as a multistate ordered character by the touch of the fingers.

3.3.3.2 Bunch

Bunch length and circumference of the bunch were measured to give a bunch length: circumference ratio. The length of the bunch was measured from the apex of the fingers of the basal node to the level of the node of the most distal hand. The circumference was taken at the widest point of the bunch.

Degree of compactness of a bunch is an important character to farmers and buyers. In this study if the average distance between each of the first three bunch nodes was less than five centimetres, the bunch was considered very compact; if between five centimetres to ten, the bunch was compact and if above ten centimetres, the bunch was lax. One clone among the East African Highland bananas is known to have no bunch nodes. The bunch has just one hand spiraling along the bunch rachis. In this case the distance between the first three turns of the hand was used to assess the degree of compactness of this bunch.

3.3.3.3 Fruits

Within the bunch, fruits vary in their arrangement, position, number in a hand, length and width, shape of apex, waxiness, anthocyanin presence, rows of ovules they contain and their shapes. In most bananas fruits are biseriately arranged in a hand, one clone is known to have biseriately arranged and fused fruits within a hand, and one other clone is known to have uniseriately arranged fruits in a hand.

Fruit position was scored as a multistate ordered character (Table 3.3). The absolute number of fingers per hand is variable. However, the number of fruits in the second hand is fairly stable. The numbers of fruits in the second hand were counted and expressed as a ratio to the number of hands in a bunch.

Unripe fruits were surveyed for skin colour, waxiness, and pulp colour. Pulp colour varied before maturity (one month after shooting) and after maturity (more than three months after shooting). Most clones have a white pulp before maturity. If the fruit is astringent, it has cream streaks across the white pulp. A few clones have cream or orange pulp before maturity. After maturity, most clones have cream or orange-brown pulp while the astringent ones have streaks, called sticky brownish excretions by Sebasigari (1987), across the coloured pulp. All young banana fruits have a degree of astringency and are bitterish. But at maturity, most fruits have almost flat or sweetish taste except the beer clones which remain astringent.

Length of fruits is important in farmers' descriptions and preferences. Measurements were made from the tip of the fruit (without floral remains) to the point where the neck of the stalk starts. The width of the fruit was measured at the widest point of the finger. Fruit shapes were scored as rounded, rectangular, triangular, gourd shaped or slender.

3.3.4 Male structures

Male inflorescence characters were scored when fingers in the bunch became negatively geotropic. This usually occurs one month after shooting and it is the time when most farmers in Uganda cut the male bud (the big purple terminal protuberance of the banana bunch) off to let the bunch fill. As flowering progresses, the volume of the male bud decreases and the shape changes because of the daily opening and falling of the bracts and flowers.

Variation is great in male inflorescence characters. The parts of the male inflorescence start below the last hand of the bunch and continue to the tip of the male bud. Only one Highland banana clone is known to have no male bud, although the condition is common among the plantains. True horn plantains have no male buds, False horn plantains have degenerating male buds while the French plantains have true male buds.

3.3.4.1 Rachis

The rachis (the long inflorescence axis which bears the fruits and the male bud) varies in position, colour, length, number of nodes present and presence or absence of persistent neuter flowers (flowers with both male and female parts which persist on the rachis below the bunch but do not develop into fruit). The rachis length was scored from the node of the last hand to the tip of the male bud. At bunch maturity, the length and number of nodes are consistent. The number of nodes was scored starting from the first empty node below the last hand of the bunch down to the last empty node above the male bud. The length and nodes of the inflorescence rachis were measured at bunch maturity.

3.3.4.2 Male bud

The male bud itself varies in many aspects which include bract arrangement, colour, shape of the bud and its apex, presence of wax and degree of waxiness. Presence of wax on male bracts was scored using the finger test. A finger print was left when a finger was rubbed against the bract if there was abundant wax. If a finger print was left, but there was no wax on the finger, wax was less abundant and this was scored as an intermediate state. If a finger print was not left at all on the bracts, then there was no wax.

3.3.4.2.1 Bracts

Bracts show variation in the way they are arranged in the bud, in their length and width, whether they curl or not after opening and colour changes inside the bract. Bract arrangement in bananas is generally convolute. This means that one margin of the bract overlaps the next one, while the other margin is overlapped by the preceding bract (Hari, 1968). There are three bracts on a male bud which are visible. Hari recommends that bract arrangement should be rejected as a character in the classification of *Musa acuminata* because it is subject to morphological changes during development. However, in this study bract imbrication was found to be a useful character in clones with a *balbisiana* genome and in a subset of clones among the Highland bananas. Hari (1968) mentioned that bract imbrication of male bud gets lost with age but the age at which this happens was never mentioned. Bracts were defined as imbricate if the apices of the lower bracts extend beyond the apices of the upper ones (Hari, 1968). Bracts of the male bud were either imbricate or not. Imbrication was prominent in male buds of clones with a *balbisiana* genome and clones of Nakitembe clone set.

The bract length was scored from the tip of the bract to the base of the bract (where the purple /blue /crimson colour stops). The width was taken at the widest point of the bract. The ratio was then defined as bract length/bract width.

3.3.4.2.2 Male flowers

Variation in male flowers was used rather than female flowers since female flowers were left to develop to maturity. The two types of flowers vary in size and function. Male flowers are smaller with a very small ovary, slender style and stigma and the anthers are morphologically well developed. Female flowers are larger, with a big well developed ovary which much exceeds the perianth in length and a big stigma. The stamens in the female flowers are reduced to staminodes. The male and female flowers also differ in behaviour. The male flowers abscise from the base of the abortive ovary and are shed whole after being exposed for about a day whereas the female flowers have no abscission layer at the base of the ovary which is therefore always persistent. Male flower characters were scored one month after shooting. A few clones were found to have tubular compound tepals which is unusual. It is usual to have an open compound tepal which is also lobed and a free single tepal which is not lobed in the banana flower. The free tepal has various shapes and the basal margins are serrated in few of the clones. The filament has a tendency to curve in some clones, and in extreme cases is shaped like a hook. This character should be observed on mature opened flowers.

3.4 Data analysis

In assessing similarities and differences among accessions, four steps were followed. These were: 1) Selection of Operational Taxonomic Units (OTUs) (objects being studied) and characters to be used in the study; 2) Standardisation of characters; 3) Measurement of overall similarities and dissimilarities between OTUs; 4) Analysis of similarities and dissimilarities between all pairs of OTUs to reveal any groupings present. The first step, which involved selection of accessions and characters for study, has been described already.

3.4.1 Standardisation of characters

Standardisation of taxonomic data is a common practice because original values are all measured on different scales. The aim is to allow each character to contribute toward the overall resemblance inversely in proportion to its variability among the entire set of OTUs used in the study. This enables a character with a small range of variation to contribute as much as another character with a large range of variation (Williams, 1976; Sneath & Sokal, 1973).

Different methods of standardisation affect characters in different ways. Some equalise the gross size of each character, others equalise variability of each character, while others do both (Sneath & Sokal, 1973). The simplest form of equalising both size and variability is by ranging, and this was the method used. Standardisation was achieved by subtracting the minimum value of each character amongst all OTUs from the value of that character for a given OTU and then the result was divided by the range for that

character over all OTUs. The general formula which was used for standardisation is given after Rohlf (1993):
y' = (y-a)/b where
y' = standardised value
y = original value of the character
a = minimum value of that character
b = range of that character

The lowest value among the OTUs would have the value 0 and the largest would have the value 1 after standardisation. The alternative method of standardisation, by standard deviation, though commonly applied in taxonomy, is best suited to normally distributed values. The characters used in this study did not have normally distributed states. This method of standardisation causes large variances for each character if used on data which are not normally distributed and this minimizes differences between groups (Sneath & Sokal, 1973; Abbot *et al.*, 1985).

3.4.2 Measures of similarity and dissimilarity

Classification (grouping of similar organisms) (Crossa *et al.*, 1995) uses measures of similarity or dissimilarity among OTUs which are calculated from measurements of the characters for each OTU. Ordination (description of spatial relationships among organisms) uses measures of similarity only.

The choice of method for measuring dissimilarity or similarity depends on the types of characters in the study. When characters occur in more than two states, as most characters in this study do, distance or correlation coefficients are the most suitable. Measurements of dissimilarity were calculated as the average taxonomic distance coefficient (Rohlf, 1993). The method involves calculating the distance between points (OTUs) whose relative positions in space are determined by their character values, to give the dissimilarity value. OTUs with least dissimilar values would be closest to each other. The formula for average taxonomic distance is shown below (Rohlf, 1993):

$$E_{ij} = \sqrt{\sum_k \frac{1}{n} (x_{ki} - x_{kj})^2}$$

where
E_{ij} = average distance between OTUs i and j.
Σ_k = the sum of values for character k.
x_{ki} = the value of character k for OTU i.
x_{kj} = the value of character k for OTU j.
n = the number of characters used in a particular comparison.

Similarity was measured by the product moment correlation coefficient (Rohlf, 1993). This measures similarity by the angular separation of lines connecting a pair of points to the origin of the character space. The correlation coefficient works out the extent to which variables are linearly related, whereas the distance coefficient assesses differences between character values (Sneath & Sokal, 1973). The formula for the correlation coefficient is shown below (Sneath & Sokal, 1973):

$$r_{ij} = \frac{\sum_{k=1}^{n} (X_{kj} - \bar{X}_j)(X_{ki} - \bar{X}_i)}{\sqrt{\sum_{k=1}^{n} (X_{kj} - \bar{X}_j)^2 \sum_{k=1}^{n} (X_{ki} - \bar{X}_i)^2}}$$

where
r_{ij} = correlation between OTUs i and j.
X_{ki} = value of character k in OTU i.
$\bar{X}_i$ = the mean of all state values for OTU i.
X_{kj} = value of character k in OTU j.
$\bar{X}_j$ = the mean of all state values for OTU j.
n = number of characters sampled.

So the different coefficients estimate different aspects of taxonomic relationships. The results are expressed in an OTU by OTU dissimilarity or similarity matrix.

3.4.3 Analysing dissimilarities and similarities

The next phase involves analysing matrices of similarity and dissimilarity. There are two major approaches to analysing similarities and dissimilarities. These are classification and ordination.

3.4.3.1 Cluster analysis

Cluster analysis is one example of classification techniques used to group items so that objects within a cluster are more similar to one another than they are to other objects in different clusters. This analysis results in a hierarchic dendrogram, produced by a sequence of fusions between OTUs or groups of OTUs.

Three widely applied agglomerative clustering techniques are employed. These are single-linkage, complete-linkage and group average clustering. Each clustering method begins by fusing the two most similar OTUs (two OTUs with least dissimilar values) to form the nucleus of a cluster. From here the methods differ in the way they form their clusters.

3.4.3.1.1 Single-linkage (nearest neighbour)

After the two most similar OTUs have clustered together, the OTUs which have the next highest similarity values (or next lowest dissimilarity values) are clustered together. If one of these OTUs is already in a cluster but the other is not, then the unplaced one joins the cluster at that similarity or dissimilarity level. Clustering continues in this way until all the OTUs are in one cluster and the dendrogram is complete. So an OTU joins a cluster by a single pairwise relation at the shortest distance or highest similarity value. This method produces long straggly clusters of OTUs, many of which may have little in common but are linked together by a chain of intermediate OTUs (Sneath & Sokal, 1973; Sokal, 1975).

3.4.3.1.2 Complete-linkage (furthest neighbour)

After the most similar OTUs have joined to form the nucleus of a cluster, other OTUs are added in a sequence determined by their distances from the furthest OTU in the cluster, with the closest (least distant) OTU joining the cluster first. Two clusters can also merge if the most distant OTUs in each cluster are closer to one another than to any OTU in any other established cluster. Clustering continues until all OTUs are in one cluster. So for an OTU to join a cluster, it must connect to all OTUs already in that cluster by a distance no greater than that between the joining OTU and the furthest OTU in the cluster. This method produces many small compact clusters and clusters join other clusters at relatively low overall similarity values (Sneath & Sokal, 1973; Sokal, 1975).

3.4.3.1.3 Group average clustering (unweighted pair group method using averages) (UPGMA)

In this method similarity (dissimilarity) between an OTU and an established cluster is the average similarity (dissimilarity) of that OTU with all OTUs in the cluster. This means that after fusion of two most similar OTUs, clustering continues between two next closest OTUs or between any unplaced OTU and the established cluster. An unplaced OTU can only join a cluster if its average similarity to all

members of the cluster is the small enough in comparison with any other pairs of unplaced OTU. The process is repeated until all clusters join into one cluster. This is an unweighted method because it gives equal weight to each OTU within a cluster. It gives only moderate type of clustering (Sneath & Sokal, 1973; Panchen, 1992).

3.4.3.2 Cophenetic correlation

Cophenetic correlation analysis is a method developed as an objective way of comparing results of different clustering methods (Sokal & Rohlf, 1962). In any given dendrogram, the cophenetic similarity between two OTUs is the similarity level of the node which links them. This value is often different from the value of the pairwise similarity (dissimilarity) coefficient between two OTUs because dendrograms inevitably distort the true relationships between individual pairs of OTUs. Cophenetic correlation coefficients attempt to test the amount of this distortion of phenetic relationships among OTUs produced by the dendrograms. Distortion can be measured by comparing a dendrogram produced by any given method with the actual similarity or dissimilarity coefficients between each pair of OTUs in the analysis.

Cophenetic correlation coefficients are calculated in the following way. The range of similarity or dissimilarity values along the axis of the dendrogram is divided into a number of equal interval classes. The number of classes will depend on the number of OTUs. These classes are given codes starting with the lowest similarity (or highest dissimilarity) value and ending with the highest similarity value (or lowest dissimilarity value). The cophenetic value of the two OTUs is the number code of the class in which their stems are connected. According to Sneath and Sokal (1973), it is no longer necessary to use codes, the actual value of each node are now used. The cophenetic correlation coefficient is calculated by calculating the product moment correlation coefficient between the original matrix value and cophenetic value. The higher the correlation, the less the distortion and probably the better the classification. There is no satisfactory statistical test for significance of the correlation, but a value of 0.8 or above is usually considered acceptable (Pankhurst, 1991). Cophenetic correlation coefficients were computed for comparison of the different clustering methods.

3.4.3.3 Principal component analysis (PCA)

Principal component analysis is an ordination technique. It produces a visual representation of the relative positions of OTUs in a space of reduced dimensions, thus indicating spatial relationships among OTUs. The position of each OTU is defined by a series of axes, each of which represents a separate character. All the OTUs could be placed along each axis according to their value for that character. If only two characters were used, a two dimensional graph would be enough to locate all OTUs. With n characters, an n-dimensional space would be required to locate all OTUs.

The line through this cloud of points that accounts for the greatest amount of variation is the first principal component. The second component is perpendicular to the first and accounts for the next greatest amount of variation. Calculation of successive components each perpendicular to the others, continues until all the variation is accounted for. The total variance accounted for by each component is called the eigenvalue. The sum of the eigenvalues will equal the original number of

characters. The proportion of variation accounted for by each principal component is expressed as the eigenvalue divided by the sum of the eigenvalues. The proportion is termed as the percentage variation of a principal component axis. Each principal component contains information from all characters but in varying proportions. These proportions form the eigenvectors (Iezzoni & Pritts, 1991). They define the relation of the principal component axis to the original data axes. Characters with large eigenvectors, either positive or negative, are considered to be large contributors to the principal component concerned. Results of principal component analysis are presented as scatter diagrams (two dimensional scatter plot). The observer assesses whether points fall into distinct clusters. PCA can be performed on two types of data matrices, a variance-covariance matrix or a correlation matrix. If the data are drawn from different kinds of measurements (lengths, counts, etc.) then a correlation matrix is probably preferable (Sneath & Sokal, 1973) and this is what was used in this study.

3.4.3.4 Discriminant analysis

Discriminant analysis is a procedure the purpose of which is to maximise the distinctness between groups already recognised on the basis of observations made on the individuals within these groups (Lebart *et al.*,1984; Grim & Yarnol, 1995).

In this study the groups of accessions were initially identified both subjectively and through cluster analysis (chapter 5). As in most classifications, some individuals do not fit well into one group or another. They are intermediate between recognised groups, causing overlap between them and creating problems of separating the groups. Cluster analysis is not good in placing these intermediate individuals. It positions them where they show similarity to only one of the groups which they resemble and not both (Pickersgill *et al.*, 1979). While the purpose of cluster analysis is to construct groups of individuals based on their overall similarity, principal component analysis provides a representation of the same data but with the number of dimensions reduced. One may then be able to distinguish groups that were not obvious in the cluster analysis. Discriminant analysis maximises differences between already recognised groups and classifies any ungrouped individuals.

Discriminant analysis is divided into two major methods: descriptive analysis and predictive or prescriptive analysis (Grim & Yarnol, 1995). Descriptive discriminant analysis involves the identification of variables or characters that best discriminate members of two or more groups (Lebart *et al.*, 1984; Manly, 1986). Predictive or prescriptive discriminant analysis allows one to predict the group to which individuals not assigned to any group may belong. Predictive discriminant analysis is sometimes called classificatory discriminant analysis (SAS Inst. Inc, 1990; Grim & Yarnol, 1995).

Although the two methods have different objectives and use different methods of analysis, many studies combine the two methods and in this study both methods were applied successively. The individuals used in the two methods are of two types. The sample of the already classified individuals is called a training set or developmental set (Grim & Yarnol, 1995). The unclassified individuals are called the hold out sample (Lebart *et al.*, 1984; Grim & Yarnol, 1995).

At the outset, data for discriminant analysis do not necessarily need to be standardised (Manly, 1986), unlike data for PCA. This is because the outcome of classificatory discriminant analysis is not affected in any way by the scaling of individual variables.

The method, however, cannot use missing values as defined in chapter 3. If missing values are retained in the analysis, all the accessions and characters with missing values become excluded from the analysis. It is therefore important to decide before commencing the analysis whether to remove the characters with missing values (if they are few) or the accessions (if they are fewer).

3.4.3.4.1 Descriptive discriminant analysis

The technique used for discriminant analyses depends on the type of data available. If the data are normally distributed a parametric method is employed and if the data are not normally distributed then a non-parametric method is used. In this study a non-parametric method was used.

3.4.3.4.1.1 Calculation of the average distances between accessions within predetermined groups

Given that descriptive discriminant analysis aims to maximise the separation of predetermined groups, the first step is to assess whether it is possible to discriminate these groups using the variables available for each individual in the different groups. The method of assessing whether variables are able to discriminate between the groups involves comparing the variability of character values between accessions in a group (which is likely to be small) and between all accessions in different groups (which is likely to be larger).

The method is best illustrated by considering the multidimensional relationships between accessions. The position the accessions occupy in space is specified by the original variables. The average distances between all accessions within each of the predetermined groups are calculated. These values provide a measure of how dispersed each group is. At this stage the clouds of points of each group in space show the approximate positions of the centroids for the different groups. As one moves from the centroids, each group becomes less dense and there is frequently an area where different groups have equal densities of points. Accessions falling in this area will be difficult to classify with certainty.

3.4.3.4.1.2 Calculation of the average distances between predetermined groups

The next stage is therefore to determine the average distances between each of the predetermined groups. The given variables are able to discriminate groups, and place unclassified accessions reliably placed in a group if the predetermined groups occupy distinct regions of the multidimensional space. F statistics are used to test the probabilities that distances between groups are significantly greater than distances within groups.

3.4.3.4.1.3 Validity of predetermined groups.

A comparison of the original classification into predetermined groups with that based on discriminant analysis is carried out in two ways:

3.4.3.4.1.3.1 Resubstitution

This process involves checking the position each accession is occupying in the dimensional space based on the original variables. The probability of an accession belonging to a group is worked out using the k nearest neighbour approach. This is a

method which gives an estimation of group membership of an individual based on k value. The k value is a preassigned figure representing a number of nearest individuals to an unclassified accession that come from any group. If the majority of these sample points nearest to an accession x are from group A then discriminant analysis assigns x to group A. If most of the k-nearest individuals to accession x do not come from the group in which an accession x was placed at the beginning of the analysis, then this disagreement is considered as a misclassification.

3.4.3.4.1.3.2 Cross validation

Cross validation involves deleting one individual from the training set sample in turn giving a new training set which is one individual smaller than the original training set and a holdout sample of one, the removed individual. The whole analysis is recomputed to determine to which group the individual that was removed belongs. In a similar manner, if it does not belong to the group to which it was originally assigned, it is considered a misclassification.

Resubstitution underestimates misclassification while crossvalidation overestimates misclassification. The actual misclassification lies between the figures of both methods and this makes it advisable to run both analyses for purposes of comparison. The proportion of correct and incorrect classification is used to calculate posterior probabilities of an accession being correctly classified.

3.4.3.4.1.4 Estimation of posterior probabilities

The posterior probability that an individual belongs to a particular group are based on k generated points, prior probabilities and the group proportions (specific group densities of different groups. The total misclassification rate which is calculated at the same time as the posterior probabilities is used to assess the effect of variations in the procedure. This is also where one tests the effect of varying the number of predetermined groups. For example several different sizes of k must be tested to find the one which gives the minimum misclassification rate using different numbers of predetermined groups. The method which gives the minimum misclassification rate gives the equations and procedure which best discriminate the predetermined groups based on the variables used.

At the end of descriptive discriminant analysis, one can identify the method that produced the lowest total misclassification rate, the equation and procedure which gave the best discrimination of groups based on the variables used. The method works better with as fewer characters as the number of individuals in the predetermined groups.

3.4.3.4.2 Classificatory discriminant analysis

Classificatory discriminant analysis uses character values which are able to maximise differences between the predetermined groups to derive an equation that will be used to predict group membership for accessions not assigned to any group (Grim & Yarnol, 1995). Based on the value k which has just been determined from descriptive discriminant analysis to produce the lowest error rate, the unclassified accessions in the holdout sample are classified. The holdout sample is classified in a similar manner to the training sample that is through resubstitution and crossvalidation.

Chapter 4

MULTIVARIATE ANALYSES ON KAWANDA BANANA GERMPLASM COLLECTION

4.1 Introduction

The history of the Kawanda collection has indicated that there has been a gradual increase in the size of the banana collection while evaluation was not being sufficiently carried out. Yet, as a collection becomes larger so the need to classify and hence order the variability present increases. Greater use of the collection can be made if there is accurate description and taxonomic identification of the accessions present and lack of these two can be an important technical constraint.

The need to describe and to classify the Kawanda germplasm collection thus became apparent and the next step was to find ways of describing and classifying the accessions. There was need initially to distinguish distinct accessions from one another and then to be able to classify the accessions into manageable groups. Groups are essential for identification and for communication especially when accessions do not belong to named cultivars or clones. Groups may also reflect genetic content and traits possessed in common, hence assist further utilisation (Baum, 1981). In order to classify the accessions in the collection, it was necessary to find ways of analysing the many different characters (Chapter 3) scored for each accession. The best way to analyse mixed data such as those recorded here is to use multivariate statistics. Classifications based on multivariate methods appear to yield more insight into phenetic relationships of cultivars and clones than most other methods (Sokal & Sneath, 1963; Baum, 1981).

A taxonomic study was therefore carried out on the Kawanda germplasm collection with three objectives:

1) to assess the usefulness of multivariate statistics in identifying and ordering variability among the accessions;
2) to determine the most useful method to separate groups of accessions;
3) to determine characters useful in recognising those groups. The study involved two separate analyses and these are reported separately. Firstly, a preliminary study was conducted using all genome groups. Secondly, the East African Highland bananas were studied alone.

4.2. Multivariate analysis of accessions of different genome groups in Kawanda germplasm collection

4.2.1 Rationale and objectives

Simmonds and Weatherup (1990b) conducted a numerical analysis using the 15 morphological characters, suggested by Simmonds and his co-workers (Simmonds & Shepherd, 1955; Simmonds, 1966; Stover & Simmonds, 1987) as useful in separating the genome groups. The analysis was based on 64 accessions representing 9 genome groups. The study produced three groups, pure *acuminata*, intermediates and predominantly or entirely pure *balbisiana*, which separated out quite clearly.

A preliminary study of accessions belonging to different genome groups was carried out on the Kawanda collection. The aims were to confirm that use of a larger and different set of characters from that used by Simmonds and Weatherup (1990b), would result in groups agreeing with the existing and widely accepted classification of Simmonds and his co-workers (Simmonds & Shepherd, 1955; Simmonds, 1966; Stover & Simmonds, 1987). The study also aimed at determining which characters are most responsible for the pattern of variation produced and whether those are the same characters that Simmonds and Shepherd (1955), Stover and Simmonds (1987), Simmonds and Weatherup (1990b) used in arriving at their classification of the genome groups.

4.2.2 Materials and methods

Seventy one accessions of *Musa* (59 AAA-EA, 5 AAA dessert, 3 AAB, 2 ABB and 2 AB) held in the Kawanda banana germplasm collection were compared for the 73 characters listed in Table 3.3 (Chapter 3). The product-moment correlation coefficient was used to assess similarity among accessions. A correlation matrix was calculated and cluster analysis carried out by the group average method. The same matrix was used in principal component analysis.

4.2.3 Results

4.2.3.1 Cluster analysis

The phenogram (Fig. 4.1) showed three main clusters. The first two clusters

(C1 to CB58 and CM4 to C4O) consisted of the East African Highland bananas. The third cluster consisted of two subclusters, one representing AAA dessert accessions (D60 to D61) and the other containing AAB, ABB and AB accessions. One would expect that the AAA dessert bananas would fuse with the East African Highland bananas first since they belong to the same genome group, AAA. However, they clustered first with accessions containing a *balbisiana* genome. It may be that the sample of accessions with a B genome used in this study had more characters in common with the dessert bananas than with the East African Highland bananas and hence these subclusters fused first.

In the phenogram one can only obtain a rough idea of the similarity of every accession with all others in the analysis. Any given pair of accessions may be more phenetically similar or more phenetically distant than is indicated by the level of

junction of stems bearing them. This is because the level of the junction represents the average resemblance of the accessions of one cluster with the accessions of the second cluster (Sokal & Rohlf, 1962). Therefore, results from cluster analysis need to be reviewed together with results from PCA for better understanding of the approximate phenetic relationship among units being studied.

4.2.3.2 Principal component analysis

The positions of accessions on the first and second components derived from the original pairwise correlations among the accessions are shown in Fig. 4.2. The first component accounted for 19% of the variation while the second component accounted for only 10% of the variation. There was a big percentage drop between components one and two in comparison with components three (7%) and four (6%). Component one was responsible for separating genome groups. The A genome triploids clustered at the left hand side while the other genome groups separated from left to right in a sequence related to the relative proportions of A and B genomes.

Component one also separated the AAA dessert accessions from the East African Highland bananas. The AAA dessert bananas occupied a position between the East African Highland bananas and the plantains. Component one did not detect clusters within the East African Highland bananas.

Component two was responsible for separating accessions within the East African Highland bananas and also separating accessions within other groups like the dessert bananas, the plantains and Ney Poovan. Most of the East African Highland banana accessions were strung out in a linear fashion along the second axis. Separation within the AAA group was obscured by the strong differences caused between the genome groups.

The grouping of accessions in PCA corresponded with that in cluster analysis in that the accessions formed clusters related to genome groups. The AAA dessert accessions shared characters with both *Musa* AAB and the East African Highland bananas and hence occupied an intermediate position between the two groups. In the phenograms produced by cluster analysis it was not obvious that they were similar to the East African Highland bananas.

On examining character loadings which caused the separation of accessions along the different axes, the following results were obtained. Of the fifteen characters used by Simmonds and Shepherd (1955), seven were scored as recommended by the two authors and all had high loadings on component one (Table 4.1). Three other characters were not scored as recommended by Simmonds and Shepherd (1955) and these had relatively low loadings on component one. Two other characters did not vary in the sample of accessions used in this study and so were not scored, while two other characters were omitted from this analysis.

In addition, five other characters which included waxy fruits, undersheath colour, compound tepal lobe colour, waxiness of male bud and petiole length/width ratio appeared to be useful in distinguishing AAA triploids from the other genome groups in this analysis. The characters separating AAAs had an average loading of 0.797. Waxy fruits were common in accessions containing a B genome. Undersheaths were commonly pink in the A genome triploids while accessions containing a B genome had green or greyed yellow undersheaths. Compound tepal lobes were orange in accessions containing a B genome and yellow in the A genome



Figure 4.1: Phenogram from UPGMA clustering of correlation coefficients between 71 accessions of *Musa* (59 AAA-EA, 5 AAA dessert, 3 AAB, 2 ABB and 2 AB) held in Kawanda banana germplasm collection. C= East African Highland bananas (AAA-EA); D= Dessert bananas (AAA); P= Plantains (AAB); S = Bluggoe (ABB); T= Ney Poovan (AB).

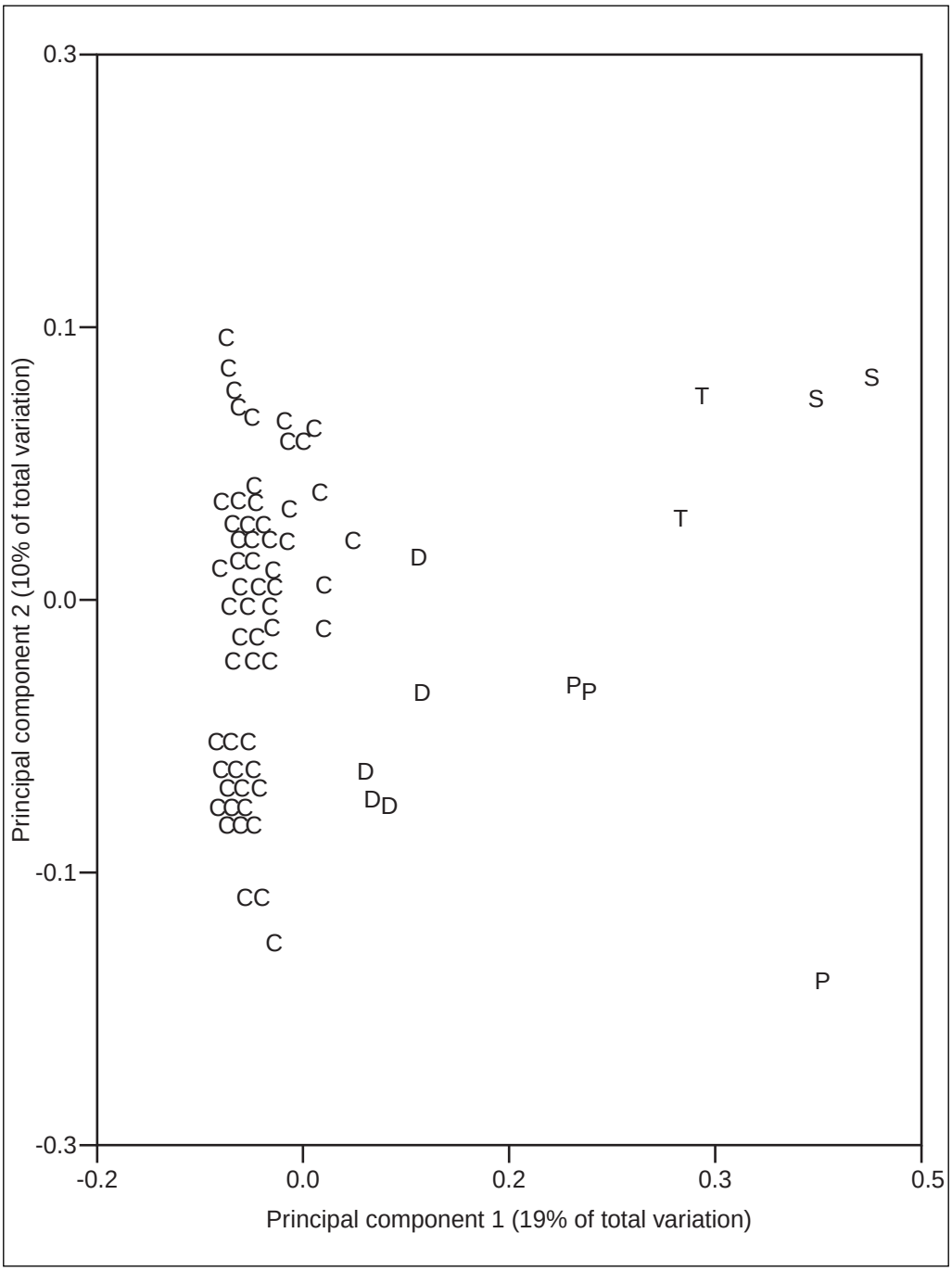


Figure 4.2: PCA showing relative positions on the first and second principal components of 71 accessions of *Musa* (59 AAA-EA, 5 AAA dessert, 3 AAB, 2 ABB, 2 AB) held in Kawanda banana germplasm collection. C= East African Highland bananas (AAA-EA); D= Dessert bananas (AAA); P=Plantains (AAB); S= Bluggoe (ABB); T= Ney Poovan (AB).

Table 4.1: Characters with a high loading on component one and useful in separating different groups in PCA

Character	Loading on component 1
1.Characters used by Simmonds and Shepherd to distinguish genome groups	
Pseudostem colour	-0.899
Petiole canal margins	-0.935
Peduncle glabrous or not	-0.905
Pedicels long or short	not scored
Ovules 2 or 4 rowed	0.630
Bract shoulder	not scored
Bract curling	0.664
Bract shape	not scored
Male bud shape (substituted for bract shape)	-0.429
Bract apex	not scored
Male bud apex (substituted for bract apex)	0.227
Bract colour	not scored
Male bud colour (substituted for bract colour)	-0.272
Colour fading	0.921
Bract scars	not scored
Free tepal of male flower	not scored
Male flower colour	0.896
Stigma colour	0.867
Mean loading (omitting substitute characters)	**0.839**
2. Characters distinguishing AAAs from *balbisiana* genome containing clones	
Waxy fruits	0.956
Undersheaths colour	0.846
Compound tepal lobe colour	0.807
Male bud waxiness	0.709
Petiole length/width ratio	0.668
Mean loading	**0.797**
3. Characters distinguishing AAA-EA from other groups	
Background colour of pseudostems	-0.846
Glossy stems	-0.749
Anther colour	0.623
Mean loading	**0.739**

triploids. Male buds were commonly waxy in accessions with the B genome and less waxy or non-waxy in A genome triploids. It is not yet clear whether these five characters (which had high loadings among the B genome containing accessions) would be useful to a larger sample in separating *Musa* AAA from the B genome containing accessions or they were only found useful because of the particular sample of accessions studied. They may need further evaluation.

Three other characters, background colour and glossiness of pseudostems and anther colour were useful in separating the East African Highland bananas from *Musa* AAA dessert bananas. These may also need further evaluation.

4.2.4 Discussion

Multivariate methods used in this study produced groups which corresponded to the genome groups of the conventional classification (Simmonds & Shepherd, 1955) and to Simmonds and Weatherup's results (1990a). PCA formed groups similar to those formed by cluster analysis. It also showed how individual accessions and groups were phenetically related to each other which cluster analysis could not. It thus gave a more accurate representation of how phenetically similar accessions were to each other, although the overall picture was very similar to that of cluster analysis.

The first component in the PCA was responsible for separating genome groups and accounted for 19% of the total variation. Of the 73 characters used in the analysis, 16 had a loading of more than 0.6 (loadings above 0.6 were considered most useful in separating groups) on component one (Table 4.1) and were therefore considered largely responsible for separating the genome groups. Of these 16, 8 had been used by Simmonds and Shepherd (1955). All characters which were not scored as recommended by Simmonds and Shepherd (1955) (Table 4.1), had low loadings (below 0.4). Although the eight characters of Simmonds and Shepherd (1955) represented a small fraction of the total of 73 characters used, they were not swamped by other characters.

Three other characters with a loading of more than 0.6 on component one had been used by other authors: waxy fruits in the recommended descriptor list as a useful character for characterisation and preliminary evaluation of different bananas (IBPGR, 1984; IPGRI, 1996,); undersheath colour by Tezenas (1983) for separating the AAAs from B genome containing clones and colour of compound tepal lobes by Sebasigari (1987), Rossel & Mbwana (1991) for separating different genome groups. Two other characters, male bud waxiness and petiole length/width ratio, had not been previously noted as characteristic of the A genome triploids. These characters should be studied in a larger sample before recommending that they form part of the extended list of characters potentially valuable in classifying bananas.

This study also revealed several other characters with a high loading on component one, useful in separating the East African Highland bananas from other groups. These were background colour and appearance (glossy) of pseudostems and pink anthers which were commonly found in the East African Highland group.

Differences within groups were not satisfactorily demonstrated by the second component. The East African Highland bananas were strung out in a linear fashion along the second axis (Fig. 4.2). The East African Highland group was therefore analysed on its own to allow any patterns of variation within this group to display themselves.

4.3. Multivariate analysis of the East African Highland bananas in Kawanda banana germplasm collection

4.3.1 Specific objectives

There were two specific objectives of this analysis. The first was to evaluate the relative merits of different multivariate methods and paying particular attention to:

i) phenetic versus subjective classification

ii) different methods of clustering

iii) clustering versus ordination techniques

The second objective was to test the subjective classification suggested at the beginning of this study.

4.3.2 Materials and methods

The 59 accessions of East African Highland bananas used in the first study were used in this analysis also. The accessions had been subjectively classified into five clone sets (Table 3.2, Chapter 3). They included 17 accessions of the Musakala clone set, 12 of the Nakitembe, 17 of the Nfuuka, 6 of the Nakabululu and 7 of the Beer clone sets.

Among the accessions were two known mutants. CM27 was known from the germplasm collection maintenance record to be a tall mutant of C27, which is commonly known as the shortest clone among the East African Highland bananas. CM32 was a dark-stemmed mutant of C32, one of the bright-coloured clones. Other accessions were claimed by farmers to be mutants. CM4 was a beer mutant of the cooking accession C4, according to the farmer from whom these two accessions were collected. Four accessions in the Nakitembe clone set were given names by farmers indicating that in their opinion they had arisen as mutants. These were Nakitembe-Nakamali (C20), Nakitembe-Nakawere (C22), Nakitembe-Omunyoro (C23) and Nakitembe-Omumyufu (C24). Nakitembe (C19) was reported by farmers to be the probable original clone although not necessarily the parent of these four accessions. Finally, C16 was said to be a mutant of C15 by the farmers although it did not share the same name.

When the analysis was restricted to the East African Highland bananas, 13 characters became invariant. These were numbers 12, 22, 13, 32, 33, 34, 37, 63, 66, 67, 68, 71, 73 (see Table 3.3, Chapter 3). The original set of 73 characters was therefore reduced to 60 characters for this study. These included 13 quantitative and 47 qualitative characters. Among the qualitative characters, there were 29 binary, 14 ordered multistate and 4 unordered multistate characters.

The product-moment correlation coefficient measures similarity and the average taxonomic distance coefficient measures distance (Chapter 3).

Three clustering methods were applied to analyse similarity and dissimilarity among the accessions. The methods were single-linkage, complete-linkage and group average clustering. PCA was performed on the correlation matrix for comparison with clustering methods.

4.3.3 Results

4.3.3.1 Cluster analyses

4.3.3.1.1 Phenetic versus subjective classification

Six phenetic classifications (Figs. 4.3, 4.4, 4.5, 4.6, 4.7, 4.8) were produced by using the three clustering methods on the two different matrices computed with the two different coefficients. The groups of accessions were similar in all 6 classifications, and also in the subjective classification. However, a few consistent differences existed between phenetic and subjective classification, suggesting modifications to the subjective classification.

4.3.3.1.1.1 Beer clone set

The Beer clone set appeared in all 6 phenograms. 6 out of the 7 accessions subjectively classified in the Beer clone set clustered together in 5 of the 6 phenograms (Table 4.2). They can therefore be considered as the heart of the clone set. In the single-linkage phenogram based on the distance coefficient (Fig. 4.4), CB58 was an outlier to all the five clone sets.

Table 4.2: Accessions which consistently clustered together in each phenogram and which belonged to the same clone set in both the phenetic and subjective classifications. Numbers in parentheses indicate the number of accessions in each clone set.

Subjective classification	Phenetic classification		Accessions belonging classification to the same clone set in all six phenograms
	Correlation coefficient	Distance coefficient	
Beer(7)	Single-link(6)	Single-link(5)	CB53, CB54,
	Complete-link(6)	Complete-link(6)	CB55, CB57
	UPGMA(6)	UPGMA(6)	CB59(5)
Musakala(17)	Single-link(13)	Single-link(12)	C1, C2, C3, C4,
	Complete-link(14)	Complete-link(12)	C5, C6, C7, C9,
	UPGMA(14)	UPGMA(13)	C10, C11, C12, C13(12)
Nakabululu(6)	Single-link(6)	Single-link(6)	C47, C48, C49,
	Complete-link(6)	Complete-link(6)	C50, C51, C52(6)
	UPGMA(6)	UPGMA(6)	
Nakitembe(12)	Single-link(8)	Single-link(7)	C19, C20, C21,
	Complete-link(7)	Complete-link(8)	C22, C23, C24,
	UPGMA(7)	UPGMA(7)	C25(7)
Nfuuka(17)	Single-link(12)	Single-link(110)	C30, C31, CM32,
	Complete-link(13)	Complete-link(9)	C35, C36, C37,
	UPGMA(12)	UPGMA(11)	C41(7)

In all phenograms, the beer mutant CM4, supposedly derived from the cooking banana C4 of the Musakala clone set, actually clustered with C18, a cooking banana of the Nakitembe clone set. In phenograms based on the distance coefficient, C18 and CM4 were outliers to all other clone sets in the analysis. When the correlation coefficient was used, CM4 and C18 were included in the Nakitembe cluster in the single-linkage (Fig. 4.3) and group average (Fig. 4.7) phenograms. However, in the complete-linkage phenogram (Fig. 4.5) CM4 and C18 combined to become part of the Musakala cluster.

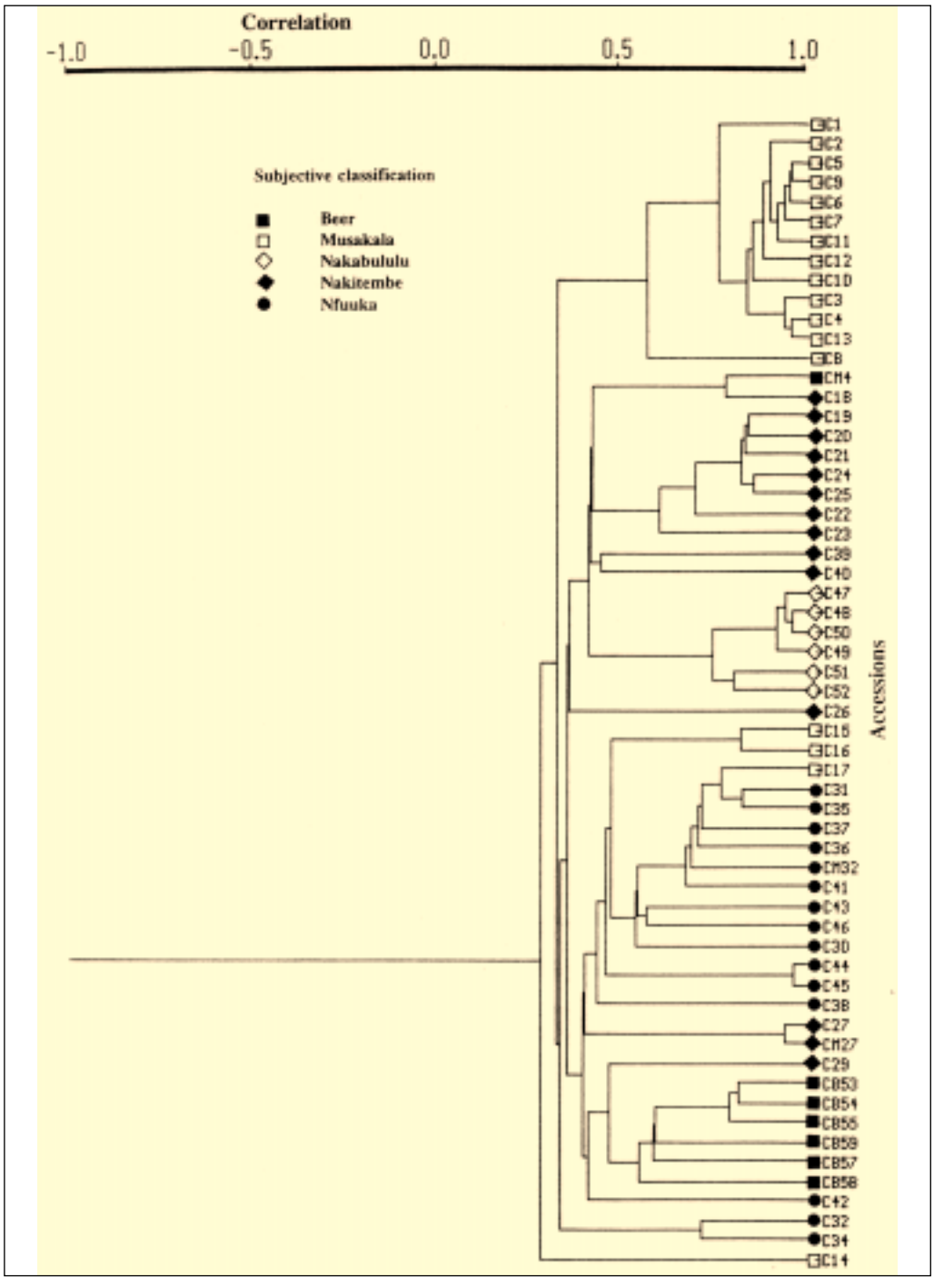


Figure 4.3: Phenogram from single-linkage clustering of correlation coefficients between 59 accessions of *Musa* AAA-EA held in the Kawanda banana germplasm collection.

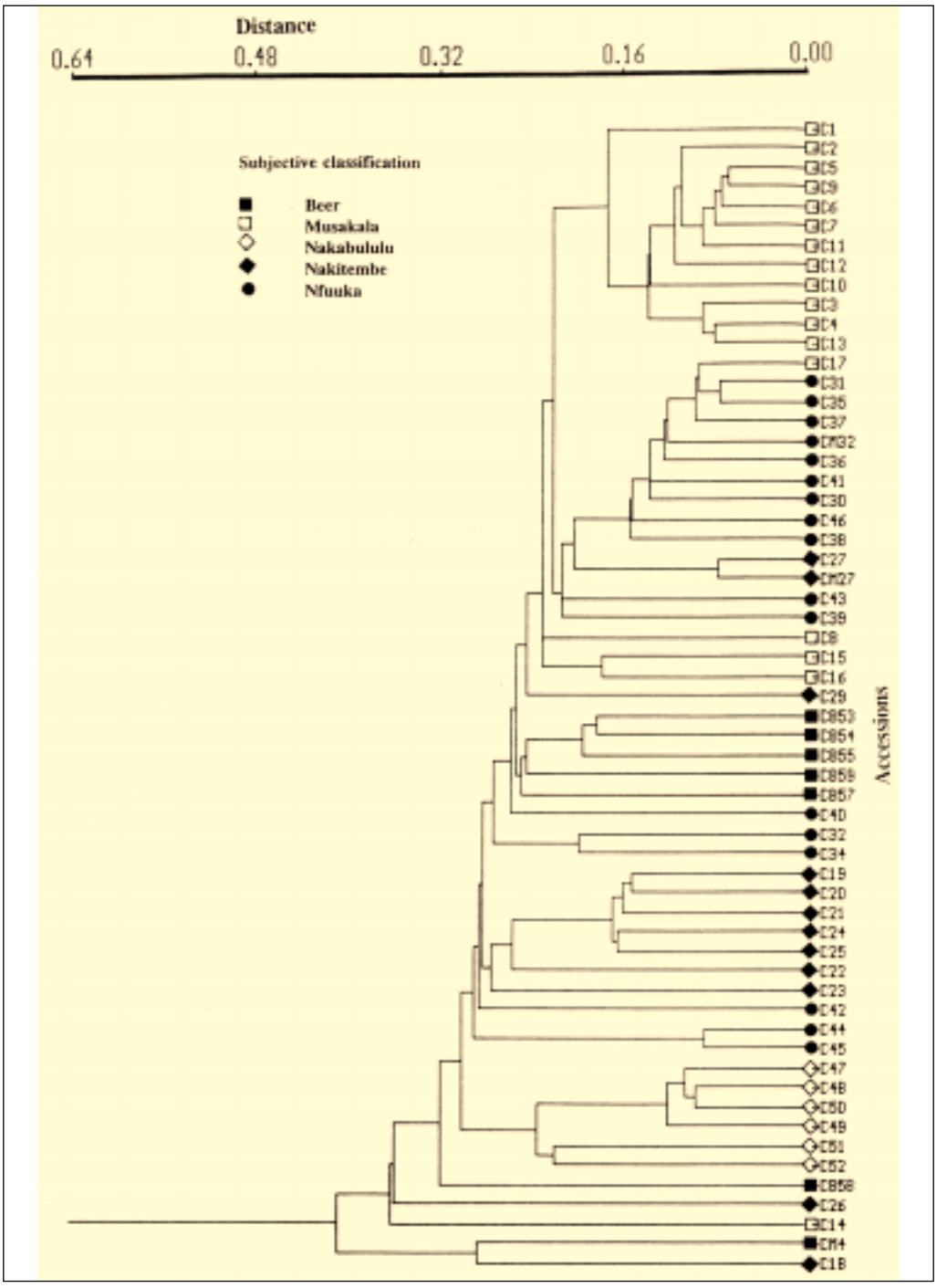


Figure 4.4: Phenogram from single-linkage clustering of distance coefficients between 59 accessions of *Musa* AAA-EA held in the Kawanda banana germplasm collection.

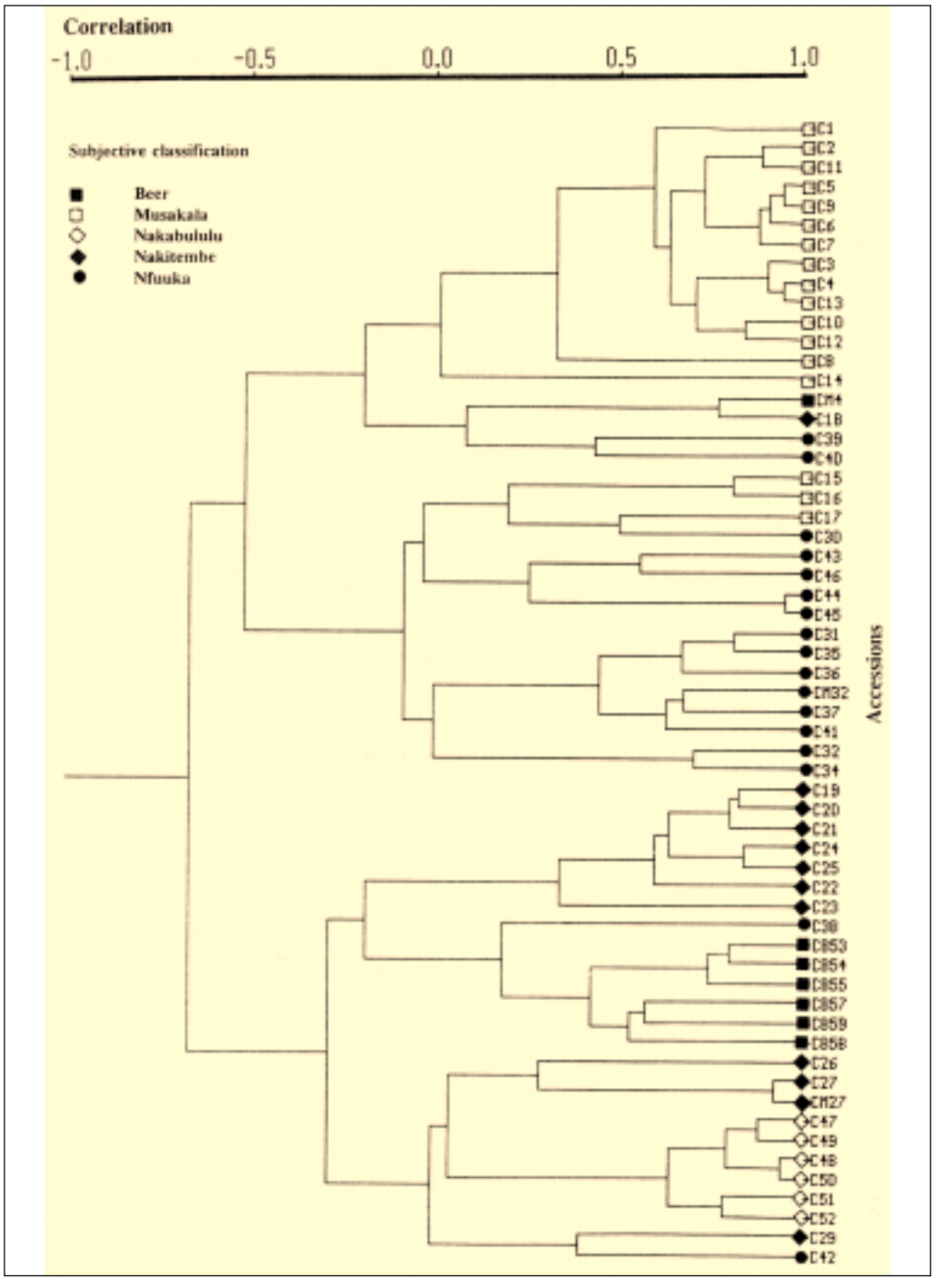


Figure 4.5: Phenogram from complete-linkage clustering of correlation coefficients between 59 accessions of *Musa* AAA-EA held in the Kawanda banana germplasm collection.

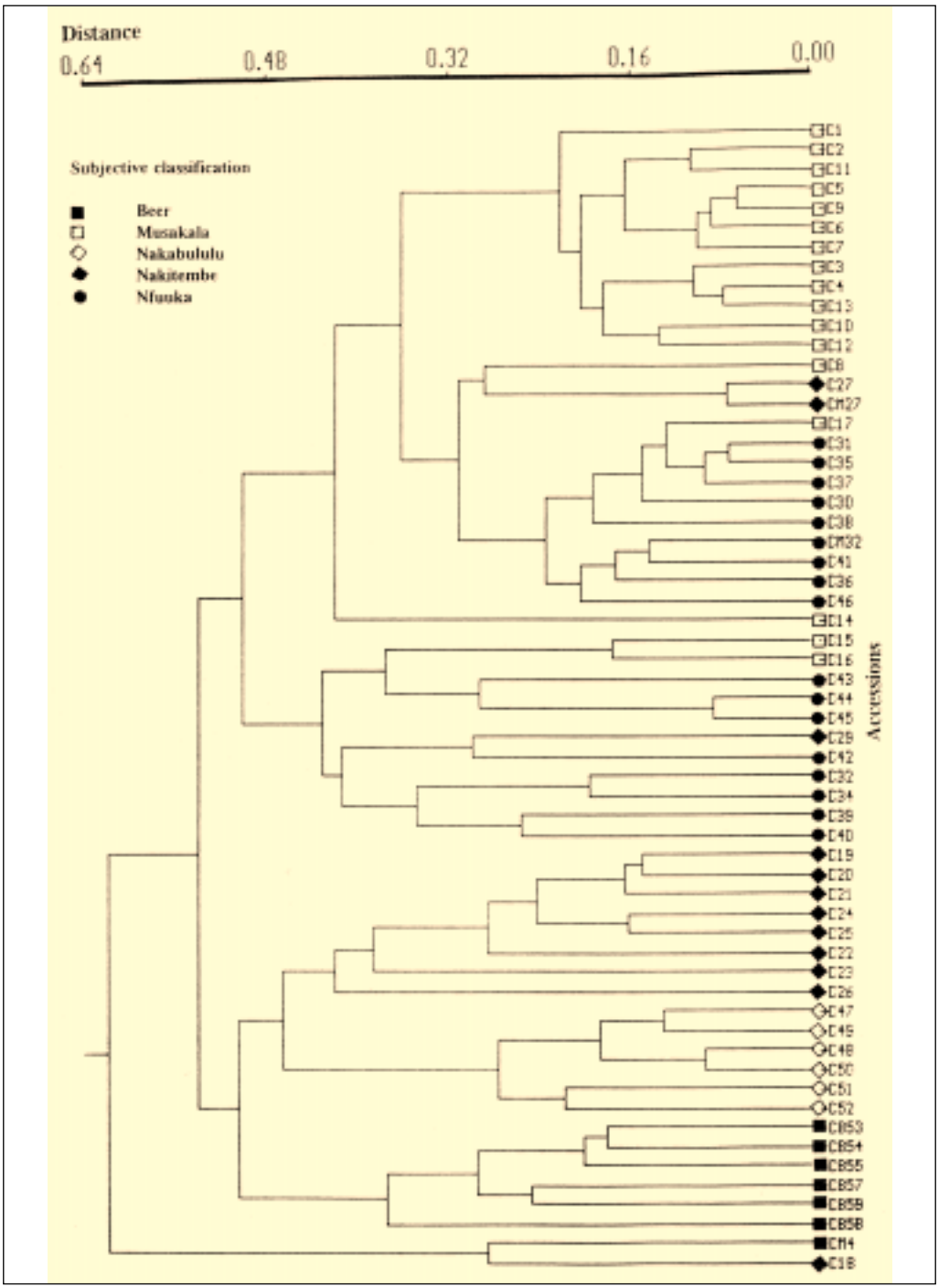


Figure 4.6: Phenogram from complete-linkage clustering of distance coefficients between 59 accessions of *Musa* AAA-EA held in the Kawanda banana germplasm collection.

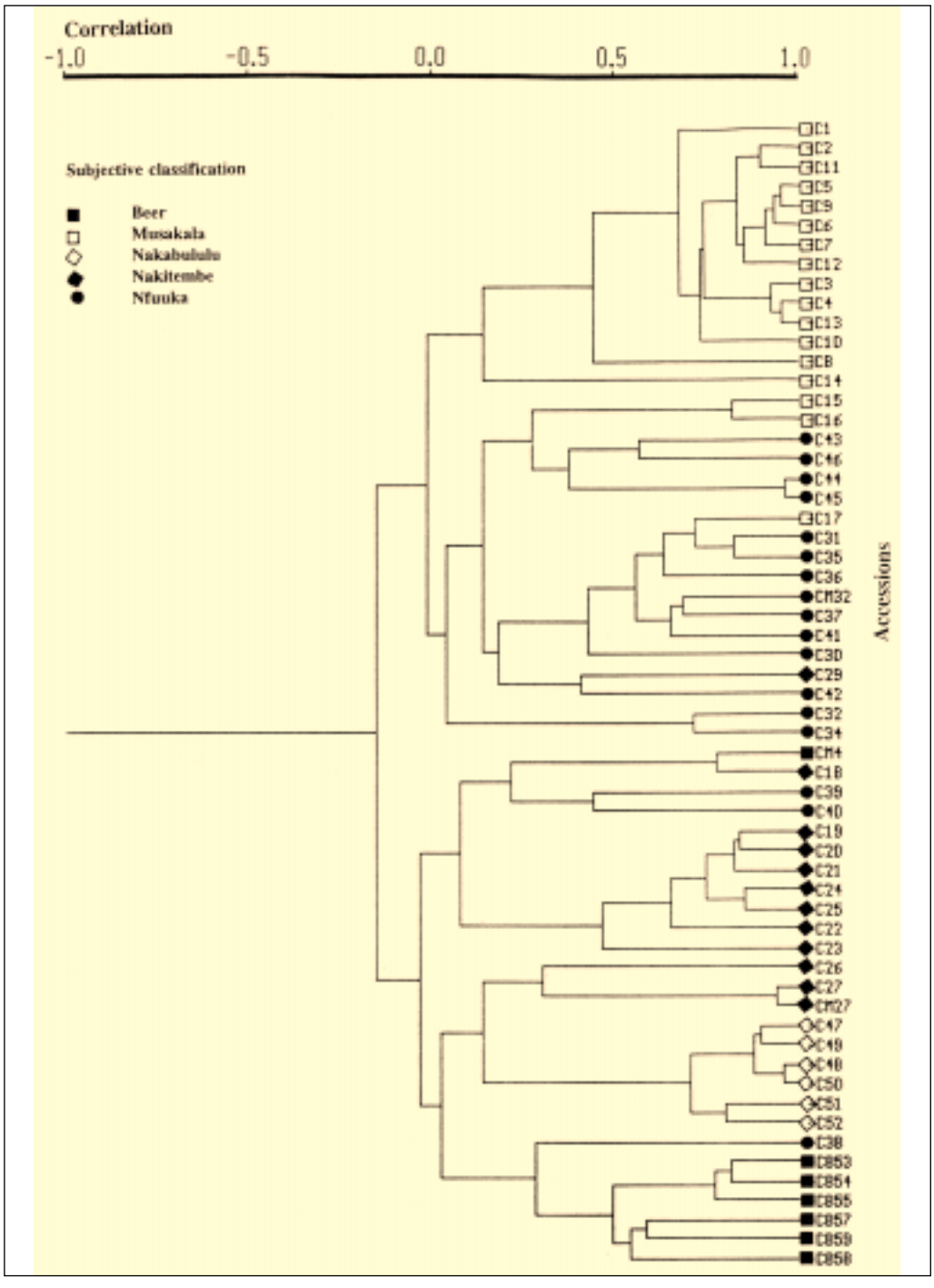


Figure 4.7: Phenogram from UPGMA clustering of correlation coefficients between 59 accessions of *Musa* AAA-EA held in the Kawanda banana germplasm collection.

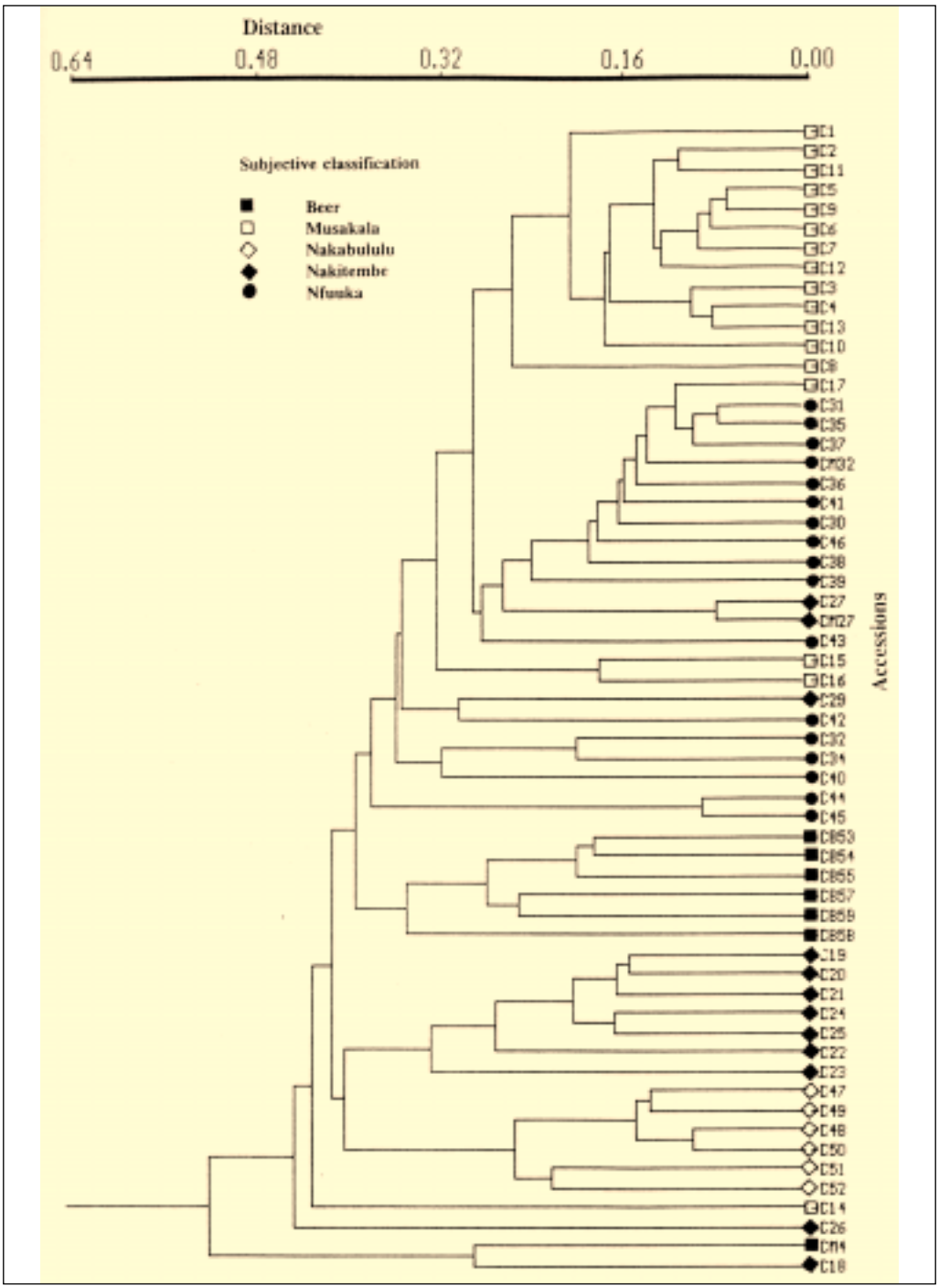


Figure 4.8: Phenogram from UPGMA clustering of distance coefficients between 59 accessions of *Musa* AAA-EA held in the Kawanda banana germplasm collection.

The position of the beer mutant CM4 outside the Beer clone set in the phenetic classifications may have several implications. Firstly, the fact that CM4 did not cluster with C4 may imply that the information given by the farmer at collection was erroneous. Secondly, some farmers claim that beer clones originate from cooking clones, while others believe that they have always existed as separate clones. If beer clones originate repeatedly and independently from different cooking clones, it may be necessary to modify the subjective classification.

4.3.3.1.1.2 Musakala clone set

The Musakala clone set of the subjective classification appeared in all 6 phenograms. 12 of the 17 accessions subjectively classified in this clone set (Table 4.2) clustered together in all the phenograms and therefore can be considered as the heart of the Musakala clone set. One accession (C17) was always grouped with the Nfuuka accessions. Three other accessions linked the Musakala clone set with the Nfuuka clone set. C15 and C16 were grouped with Nfuuka in all 3 phenograms based on the correlation coefficient, while single linkage and group average clustering of the matrix of distance coefficients (Figs. 4.4 and 4.8) placed them as outliers to the combined Musakala-Nfuuka cluster. When the correlation coefficient was used, C8 was included in the Musakala clone set, but as one of the two most dissimilar members of the group. When the distance coefficient was used, C8 clustered with Nfuuka (Fig. 4.6) or with the combined Musakala-Nfuuka cluster (Fig. 4.4), or as a dissimilar member of the Musakala clone set (Fig. 4.8). C14 appeared as an outlier to all the East African Highland bananas in both phenograms clustered by single-linkage. Its highest correlation coefficient (0.273) was with C1 (Musakala clone set) while its lowest distance coefficient (0.362) was with C35 (Nfuuka clone set).

The 6 phenograms therefore all supported the recognition of the Musakala clone set, but suggested that it may intergrade with Nfuuka. C15 and C16, and perhaps C8, are examples of intermediates between the two groups. C17 should probably be reclassified as Nfuuka while the position of C14 may need to be reviewed after PCA results.

4.3.3.1.1.3 Nakabululu clone set

A cluster corresponding to the Nakabululu clone set appeared in all phenograms and had exactly the same membership as in the subjective classification (Table 4.2). This was the clone set with the smallest number of accessions and the greatest homogeneity within the clone set.

4.3.3.1.1.4 Nakitembe clone set

The Nakitembe clone set appeared in all the 6 phenograms. 7 out of the 12 accessions subjectively classified as Nakitembe clustered together in all phenograms. These constitute the heart of the clone set (Table 4.2).

Of the 5 other accessions, C18 appeared to be the most dissimilar member of the group. Its lowest distance and highest correlation coefficients were with CM4 which as already discussed is reportedly a beer mutant derived from C4 (Musakala clone set).

C26 appeared as an outlier to all clone sets in single-linkage (Fig. 4.4) and group average (Fig. 4.8) clustering of the matrix based on distance coefficients, while complete-linkage (Fig. 4.6) clustering placed it as an outlier to the main cluster of Nakitembe accessions. However, when the correlation coefficient was used, C26

appeared closer to the Nakabululu clone set than to Nakitembe. The accession was an outlier to the combined Nakabululu and Nakitembe clusters in single-linkage (Fig. 4.3) clustering. Together with C27 and CM27, C26 joined the Nakabululu cluster in both complete-linkage (Fig. 4.5) and group average (Fig. 4.7) clustering. In single-linkage (Fig. 4.3) clustering of the matrix of correlation coefficients, C27 and CM27 joined the Nfuuka cluster. They also clustered with Nfuuka in the 3 phenograms based on distance coefficients (Figs. 4.4, 4.6, 4.8).

C29 was placed in the Nfuuka cluster in three phenograms (Figs. 4.6, 4.7, 4.8) but joins the combined Musakala-Nfuuka cluster in one phenogram (Fig.4.4). It was an outlier to the beer cluster in the single-link phenogram (Fig. 4.3) based on the correlation matrix and an outlier to Nakabululu cluster in the complete linkage (Fig. 4.5) phenogram also based on the correlation matrix. This is an accession which seems to be phenetically similar to several clone sets but may probably belong to Nfuuka clone set.

The analysis suggested that Nakitembe may intergrade with the Nakabululu group, C26 being an example of an intermediate accession. C27, CM27 and C29 should probably be reclassified as Nfuuka accessions. The position of C18 may need further consideration when a bigger sample has been analysed.

4.3.3.1.1.5 Nfuuka clone set

The Nfuuka clone set had the most accessions and was the most heterogeneous. All the 6 phenograms detected this group. 7 accessions (C30, C31, CM32, C35, C36, C37, C41) out of 17 subjectively classified in the Nfuuka clone set clustered together in all the 6 phenograms and can be considered the heart of the clone set (Table 4.2). The 10 other accessions (C32, C34, C38, C39, C40, C42, C43, C44, C45, C46), sometimes formed a second subcluster but sometimes would split up and join other clusters. C32 was clustered at high levels of similarity with C34 most of the time; C38 with Beer accessions, C39 with C40; C42 with C29 (Nakitembe clone set) and C43 together with C44, C45, and C46.

Nfuuka accessions need further study by PCA to understand the relationships within the clone set since there is much heterogeneity within the clone set and to establish the boundaries between Nfuuka and other clone sets.

4.3.3.1.1.6 Mutants

The Highland bananas are believed to have derived from a single clone but were greatly diversified by somatic mutations (Tothill, 1970) and they continue to do so. This means that each clone is either an old or new mutant. The old mutants may be different from the original clone but new mutants may be similar to the original parent depending on the type of change. Based on the correlation and distance coeffients, the mutants under the study were either similar to the original parent or they were very different (Table 4.3).

Of the two mutants which arose within the collection, CM27 (the tall mutant of C27) was linked to C27 in all phenograms indicating a close phenetic relationship between the two. CM27 shared almost all characters with its parent except for quantitative characters connected with height and girth of the pseudostem, ratios of length and width of parts of the vegetative and female inflorescence structures (Table 4.4). This is a recent mutant.

Table 4.3: Mutants and their estimated phenetic relationships with their presumed parents.

Mutant	Most similar accession on basis of:		Similarity with presumed parent on basis of:	
	Distance coefficient	Correlation coefficient	Distance coefficient	Correlation coefficient
1. Mutants observed to have arisen in the collections				
CM27	C27 = 0.078	C27 = 0.933	0.078	0.933
CM32	C37=0.121	C37=0.680	0.306	0.054
2. Accessions farmer claimed to be a mutant at time of collection				
CM4	C18=0.287	C18=0.770	0.452	0.238
C16	C15=0.179	C15=0.811	0.179	0.811
3. Clones supposed to be mutants because they share the name Nakitembe				
C20	C19=0.152	C19=0.832	0.152	0.832
C22	C24=0.256	C21=0.685	0.273	0.634
C23	C20=0.274	C20=0.591	0.286	0.548
C24	C25=0.164	C25=0.843	0.179	0.777

Table 4.4: Characters differentiating known mutants from their parent accessions

Character	State in parent	State in mutant
a) C27 and CM27	**C27**	**CM27**
Pseudostem height/girth ratio	3.6	3.8
Petiole length/width ratio	2.0	1.8
Distance between petiole bases	7.2	11.9
Leaf length/girth ratio	2.1	2.3
Peduncle length/girth ratio	1.7	2.4
Bunch length/width ratio	0.4	0.5
Finger/cluster ratio	2.6	2.3
Fruit width/pericarp thickness ratio	11	10.4
Fruit length/width ratio	3.9	3.5
Rachis nodes	32	22
Male flower/ovary ratio	2.5	3.1
b) C32 and CM32	**C32**	**CM32**
Pseudostem background appearance	bright coloured	dark coloured
Pseudostem undersheath colour	yellow-green	masked by red
Undersheath anthocyanin	absent	present
Pseudostem blotches	absent	present
Blotch colour	non applicable	black
Petiole length/width ratio	2.9	2.5
Distance between petiole bases	15	11
Peduncle length/width ratio	2.3	2.1
Bunch length/circumference ratio	0.4	0.7
Finger/cluster ratio	2.6	1.9
Fruit length/width ratio	3.7	3.6
Rachis length	65	84
Rachis nodes	41	89
Ovary/flower ratio	3.1	4.0

CM32, the dark stemmed mutant of C32 did not show any close phenetic relationship with C32. It was more similar to C37. The original data showed that CM32 differed from C32 in pseudostem appearance, pseudostem undersheath colour and anthocyanin presence, pseudostem blotches and their colour, the bunch length/circumference ratio and the length/width ratio of fruits (Table 4.4). C32 was maintained in a different strip from CM32, where the soils were hard and stony. The quantitative characters by which CM32 differed from C32 could have been affected by the growing conditions. Farmers have explained that C32 commonly changes to a black or brown stemmed mutant, at times accompanied by changes in the bunch or in the taste of the food cooked from the fruits. Farmers have also claimed that this accession can change to C31, C34, C37, C44 and C45. After such a change farmers call it Nfuuka (meaning I keep changing). A few farmers use the name Namwezi Black, indicating Namwezi white has changed to black. The name of the original parent is commonly retained if the change is small i.e. if one can still recognise characters which show that it is Namwezi.

C16 is an accession claimed by farmers to be a mutant of C15. The similarity and dissimilarity matrices showed C16 to be more similar to C15 than to any other accession in the analyses (Table 4.3). They share most of their characters except colour differences on upper sheaths, petioles and midribs (Table 4.5). Farmers also indicate that they are both slow to mature (Siira means “go slowly”) and their fruit is hard after cooking.

Differences between CM4 and C4 have been noted in section 4.3.3.1.1. Tables 4.3 and 4.5 demonstrate that these are large differences. The farmer’s statement that C4 gave rise to CM4 was probabaly not correct since CM4 differs from C4 in more than three-quarters of the qualitative characters and is more similar to C18 than to C4.

Accessions C20, C22, C23, C24, which shared the first part of their name with Nakitembe (C19) and which are probably mutants of Nakitembe, clustered with C19 and formed the heart of the Nakitembe clone set. However, according to both the correlation and distance matrices C20 and C24 were closer to C19 than C22 and C23. According to farmers C20 differs from C19 by having a slender pseudostem and a smaller bunch. Hence the second part of the name of C20 means “small Nakitembe”. C20 differed from C19 by 6 quantitative characters but no qualitative characters (Table 4.5).

C24 is similar to C19 but has red upper sheaths, petioles and midribs. However, C22 differs from C19 by having watery green upper sheaths, petioles and midribs; it has a non waxy bright crimson male bud and a fresh persistent style on fruits (Table 4.5). Bunches and fruits are smaller in C22 than in C19. C23 is also significantly different. It has a cylindrical bunch while all other Nakitembe accessions have a rectangular bunch. Fruits of C23 are rounded, blunt and arranged at right angles to the axis within the bunch. C19 has rectangular bottle-necked fruits borne at an angle of 45° to the bunch axis.

4.3.3.2 Principal component analysis

Table 4.6 shows the first 7 principal components and the percentage variation extracted by each. Together these first 7 components accounted for 53% of the total variation. The first component accounted for only 13% of the variation while

the succeeding components accounted for successively less of the remaining

Table 4.5: Characters differentiating reported mutants from their parents.

Character	State in parent	State in mutant
a) C15 and C16	**C15**	**C16**
Plant height/girth ratio	4.8	4.5
Petiole background colour	masked by red	green
Petiole anthocyanin	on adaxial strie	in marginal areas
Petiole length/width ratio	2.9	2.4
Leaf length/width ratio	3.2	2.9
Peduncle length/girth ratio	2.2	2.1
Bunch length/circumference ratio	0.6	0.5
Finger/cluster ratio	2.3	2.1
Fruit width/pericarp ratio	11	10.3
Fruit length width ratio	4.2	4.5
Rachis length	55	66
Rachis nodes	72	45
Bract length/width	1.3	1.4
Ovary ratio	3.0	4.0
b) C4 and CM4	**C4**	**CM4**
Plant height/girth ratio	4.2	4.4
Distance between petiole bases	17	18
Leaf length/width ratio	2.5	2.9
Peduncle length/girth ratio	1.9	2.4
Bunch length/circumference ratio	0.4	0.3
Fruit length/width ratio	4.2	4.1
Persistent style	non-persistent	persistent
Type of persistent style	non-applicable	fresh style
Persistent stamen	non-persistent	persistent
Rachis length	72	76
Persistent neuter flowers	non-persistent	persistent
Rachis nodes	28	81
Male bud anthocyanin	present	absent
Male bud anthocyanin colour	purplish blue	non-applicable
Male bud waxiness	intermediate	non-waxy
Bract imbrication	not imbricate	imbricate
Ovary ratio	4.1	2.9
Free tepal serrated	sonate at margins	non serrated

c) Nakitembe complex	C19 (parent)	C20	C22	C23	C24
Plant height/girth ratio	4.9	4.7	4.8	4.7	4.5
Petiole background colour	green	green	watery green	green	masked by red
Petiole anthocyanin	absent	absent	absent	absent	present
Petiole length/girth ratio	2.8	2.5	2.8	2.6	3.2
Distance between petiole	13	12	16	15	13
Leaf length/width ratio	3.3	3.1	3.7	2.9	3.1
Peduncle length/girth	2.4	2.2	2.5	2.0	2.2
Bunch ratio	0.4	0.6	0.5	0.7	0.5
Bunch shape	rectangular	rectangular	rectangular	cylindrical	rectangular
Finger/cluster ratio	2.0	1.7	2.3	2.4	2.3
Fruit position in the bunch	positively geotropic	positively geotropic	positively geotropic	none	positively geotropic
Fruit width/pericarp ratio	11.3	13.4	11.3	11.7	11.0
Fruit apex	intermediate	intermediate	intermediate	intermediate	intermediate
Type of persistentstyle	dry	dry	dry	fresh	dry
Persistent stamens	absent	absent	absent	present	absent
Fruit length/width ratio	3.8	3.4	3.7	3.9	3.8
Fruitshape	rectangular	rectangular	rectangular	round	rectangular
Rachis position	oblique	oblique	oblique	pendulous	oblique
Rachis length	47	59	59	52	58
Rachis nodes	33	29	21	80	21
Bract ratio	1.2	1.3	1.3	1.6	1.2

variation. So it was necessary to examine a number of components to determine those bringing out differences among the East African Highland bananas most effectively.

The positions of accessions on the first, second, third and fourth principal component axes derived from the original pairwise correlations among accessions are shown in Figs. 4.9, 4.10, 4.11. These plots showed that accessions fell into five clone sets similar to those of the subjective classification. Lines were drawn around the accessions which clustered together in each phenogram in a position which agreed with their subjective grouping. Accessions which did not cluster as expected on the basis of the classification or inconsistently placed with regard to the six phenograms are marked with their individual codes. The majority of these accessions occupy intermediate positions relative to the five clone sets so far recognised. However, CM4 and C18 could represent a new clone set.

PCA has therefore shown that accessions inconsistently placed in the phenograms are mostly intermediate between the recognised clone sets.

Table 4.6: The first 7 principal components showing their percentage contribution to the total variation.

Components	Percentage variation explained by each component
1	13%
2	9%
3	9%
4	7%
5	6%
6	5%
7	5%
Total	**53%**

Component one separated four clone sets of accessions; Musakala, Nfuuka, Nakitembe, (Beer overlaps Nfuuka and Nakitembe) and Nakabululu clone sets.

Component two separated the Nakabululu clone set further from the rest, and separated two accessions, C18 and CM4, from all others in the analysis. Component three was responsible for separating the Nakitembe clone set plus three accessions of the Nfuuka clone set (C40, C44, C45) and also CM4 and C18 from the rest. Component four separated the Beer clone set from the rest and it also separated C32 (subjectively placed in the Nfuuka clone set) from all other accessions.

Of the 60 characters used in the analysis, 11 had a loading of more than 0.5 on the first component (Table 4.7). 6 of these had been used in arriving at the subjective classification (Table 4.7). 7 characters had negative loadings above 0.5 and were useful in defining the Musakala clone set. These were leaf tip twisting, bunch orientation, bunch shape, fruit apices, fruit length/width ratio and rachis position (Table 4.7). Leaf tip twisting and bunch shape were not used in the subjective definition of the Musakala clone set.

Four characters had high positive loadings on component one and were useful in distinguishing Nakabululu clone set. These included bunch compactness, fruit position in the bunch and pulp colour both before and after maturity of the fruit. 3 of these characters were used in defining the Nakabululu clone set. Fruit position also appears useful for this purpose.

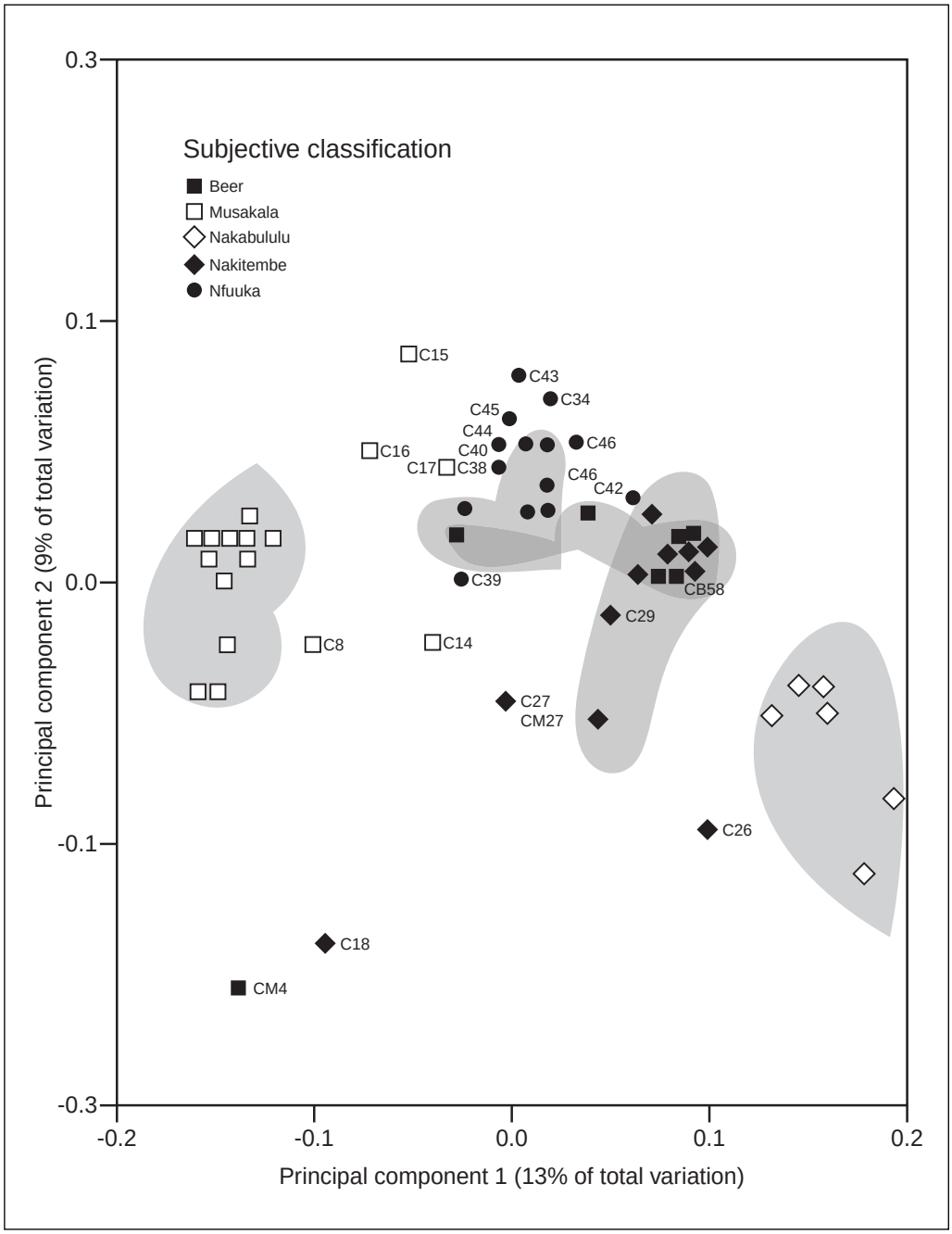


Figure 4.9: Positions of 59 accessions of *Musa* AAA-EA held in Kawanda banana germplasm collection with respect to 1st and 2nd principal components. Lines drawn around some of the symbols show accessions which clustered together in each phenogram in a position which agreed with their subjective grouping. The labelled accessions were inconsistent in their positions with regard to different phenograms and the subjective grouping.

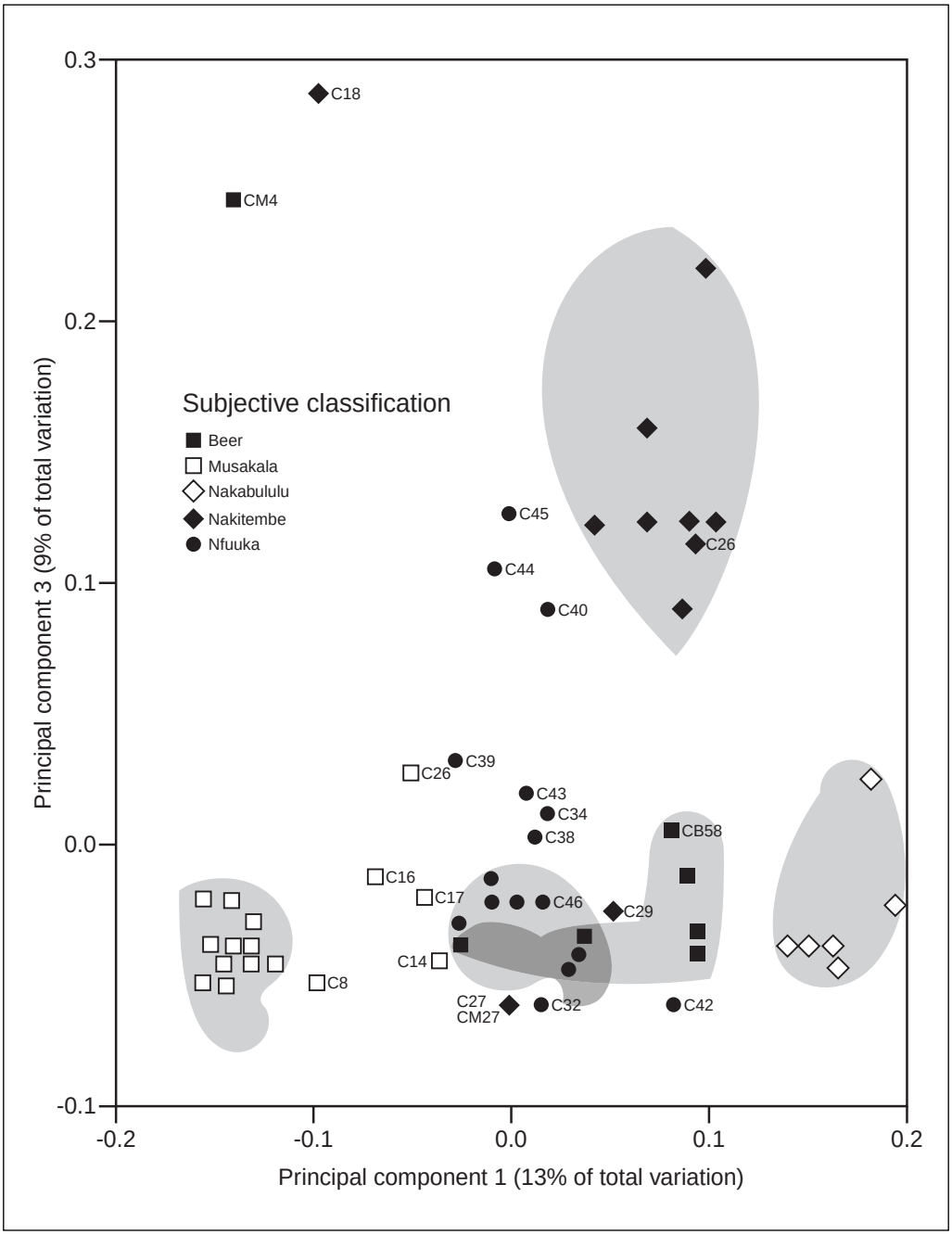


Figure 4.10: Positions of 59 accessions of *Musa* AAA-EA held in Kawanda banana germplasm with respect to 1st and 3rd principal components. Lines drawn around some of the symbols show accessions which clustered together in each phenogram in a position which agreed with their subjective grouping. The labelled accessions were inconsistent in their positions with regard to different phenograms and the subjective grouping.

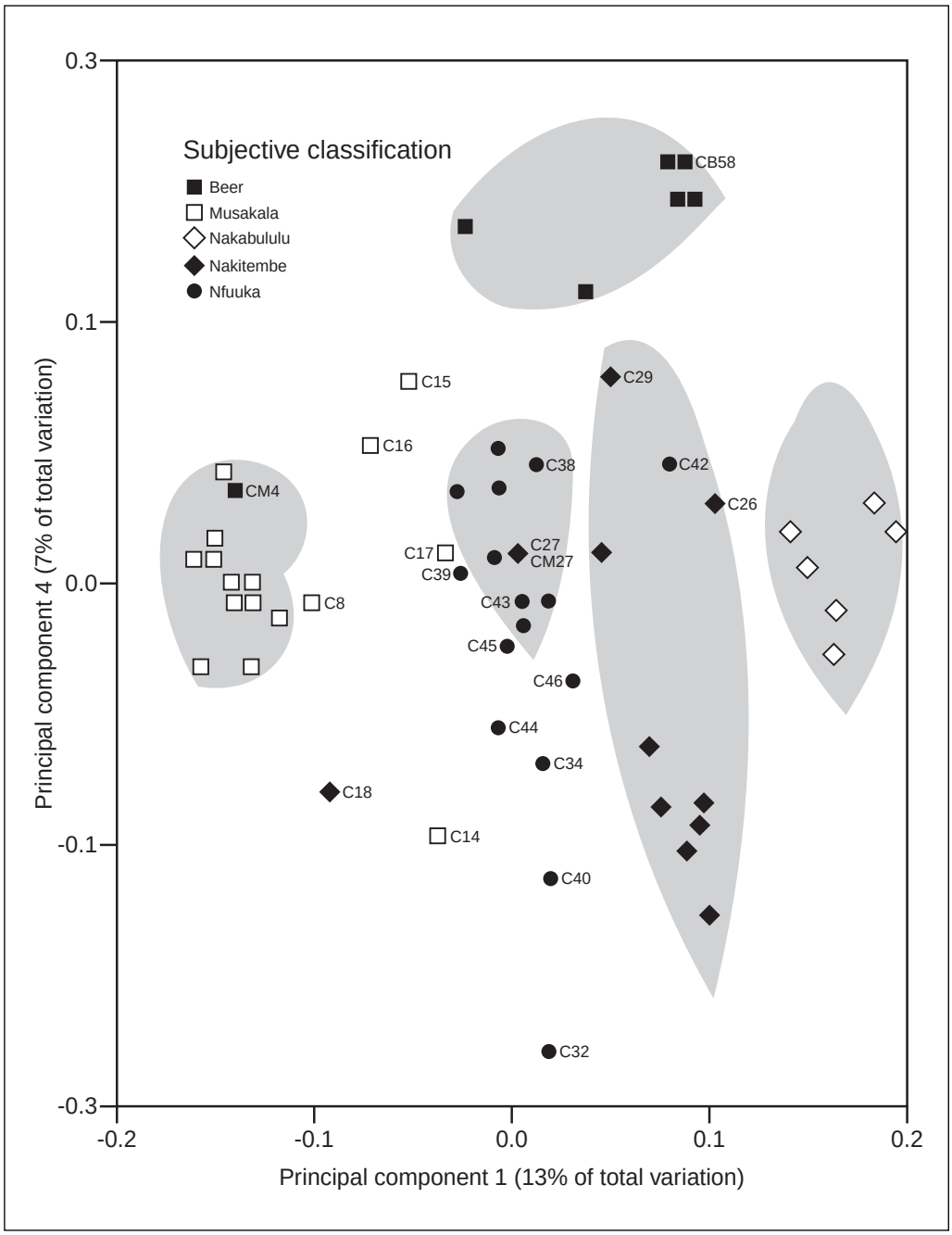


Figure 4.11: Positions of 59 accessions of *Musa* AAA-EA held in Kawanda banana germplasm collection with respect to 1st and 4th principal components. Lines drawn around some of the symbols show accessions which clustered together in each phenogram in a position which agreed with their subjective grouping. The labelled accessions were inconsistent in their positions with regard to different phenograms and the subjective grouping.

Table 4.7: Characters with loadings greater than 0.5 (in bold) on principal component one, two, three or four.

Character	PC 1	PC 2	PC3	PC4
Plant height/girth ratio	0.095	**0.715**	0.333	0.044
Undersheath colour	0.050	0.412	**0.597**	**0.849**
Petiole length/width	0.216	**0.659**	0.360	0.210
Leaf tip	**-0.750**	-0.186	-0.135	0.027
*Bunch orientation	**-0.823**	0.033	0.003	0.104
Bunch shape	**-0.769**	-0.193	0.066	0.128
*Bunch compactness	**0.820**	-0.054	-0.129	0.152
Fruit position in a bunch	**0.627**	-0.254	-0.402	0.103
*Pulp colour at immaturity	**0.560**	-0.309	0.268	0.143
*Pulp colour at maturity	**0.543**	-0.188	0.299	0.418
Pulp with sticky excretions	0.107	-0.102	0.005	**0.516**
Pulp taste	0.211	0.083	-0.155	**0.504**
*Fruit apex	**-0.803**	-0.096	0.019	-0.246
Type of perst. style	-0.261	**-0.963**	0.026	-0.224
Persistent stamen	0.167	**-0.609**	0.329	0.386
*Fruit length/width ratio	**-0.757**	0.155	0.239	0.200
Fruit shape	**-0.876**	-0.182	-0.004	0.244
*Infl. rachis position	**-0.745**	-0.331	-0.086	-0.131
*Persistent neuter flowers	0.257	-0.381	**0.615**	0.215
Rachis nodes	-0.158	0.019	0.252	**0.534**
Male bud anthocyanin	0.203	0.340	**-0.514**	0.078
*Male bud shape	0.227	**-0.798**	-0.106	0.027
Male bud waxiness	0.098	0.281	**-0.654**	0.172
Male bud apex	0.318	**-0.774**	-0.115	0.042
*Bract imbrication	0.161	-0.233	**0.703**	-0.203
Free tepal serrated	-0.206	-0.193	**0.588**	-0.064

*** characters used in subjective classification**

Six characters accounted for much of the variation contributed by the second component. Of the 6, only male bud shape was used in the subjective classification.

The remaining two characters, relating to the persistence and nature of the stamens and style, distinguished CM4 and C18 from all other accesions.

Five characters contributed largely to component three which separated the Nakitembe clone set from other clone sets and also separated three accessions of the Nfuuka clone set (C40, C44, C45) and CM4 and C18 from the other accessions. Only two of these characters were used in the subjective classification: persistent neuter flowers and bract imbrication. The other 3 characters serrated free tepals, male bud waxiness and anthocyanin in male bracts separated a few accessions but were not characteristic of major clusters of accessions.

Two characters, pulp with sticky excretions and pulp taste had loadings above 0.5 on component four and were responsible for the separation of beer accessions from other groups. These two characters were important in the subjective definition of the Beer clone set. Undersheath colour also had a high loading and contributed to the separation of C32 from all other accessions, since C32 is the only accession whose undersheath is bright green.

Twenty six characters altogether have contributed to the separation of cluster of accessions within the Highland bananas. Ten of these had been used at the outset in the subjective grouping but the other sixteen had not appeared particularly valuable.

4.3.3.3 Discussion

Cluster analysis has become a popular method among scientists engaged in classifying items. Commonly clusters represent disjoint sets of individuals in a phenogram and it is hoped that these sets correspond to marked features of the sample (Gower, 1967). It may be very difficult to judge the relative merits and demerits of different methods of clustering. Sneath and Sokal (1973) state that the structure and composition of clusters are important criteria for comparing the different methods of clustering. In this analysis we have seen that different methods of cluster analysis of the same sample produced clusters of different sizes and compositions.

Single-linkage clustering produced phenograms which had much chaining, so that clusters were not well delineated. This agrees with the experience of others (Sneath & Sokal, 1973; Abbot *et al.*, 1985; Peeters & Martinelli, 1989).

Complete-linkage clustering produced phenograms with well spaced, well delineated, and tight clusters which joined other clusters at relatively low overall similarity values (Panchen, 1992).

Group average clustering produced well defined and fairly compact clusters. The majority of clusters corresponded with the clone sets of the subjective classification. Cophenetic correlations were computed to test the goodness of fit of the phenograms produced by the different methods of cluster analysis to the original data in the similarity or dissimilarity matrices (Table 4.8). Cophenetic correlations above 0.8 indicate good fit to the original matrix and those below indicate a poor fit. Cophenetic correlations for group average clustering were higher than those for other methods, which was to be expected according to those who have used the method (Sneath, 1969; Boyce, 1969; Sneath & Sokal, 1973). This method was therefore in subsequent analyses. Single-linkage clustering produced low cophenetic values and results were in agreement with those who have used this method before (Sneath, 1969; Boyce, 1969).

Table 4.8: Cophenetic correlation coefficients for the six phenograms.

Clustering method	Average distance coefficient	Product-moment correlation coefficient
Single-linkage	0.86	0.79
Complete-linkage	0.83	0.80
Group average	0.89	0.83

As discussed earlier (chapter 3), the two coefficients estimate different aspects of taxonomic relationship and phenograms based on the different coefficients may therefore differ. The two coefficients were indeed found to give different pictures of resemblances among the accessions section (4.3.3.1). A further look at their perfomance with the larger sample is considered in the next phase.

In general, PCA results agreed with results obtained by cluster analysis. There was agreement between the subjective classification, the phenetic classification and PCA with regard to the positions of the majority of accessions in each subjective

clone set. A number of accessions were, however, not consistently placed in the different phenograms. These accessions appeared intermediate between one or more clone sets in PCA. A few of these may need reclassifying in other clone sets, while others may belong to clone sets not recognised in the subjective classification. Others looked to be intermediate between several clone sets.

Twenty six characters had a loading of more than 0.5 on at least one of the first four axes of PCA. Some of these appear useful in distinguishing clone sets within the Highland bananas. These characters need to be evaluated on a larger sample to confirm their usefulness in distinguishing clone sets of the East African Highland bananas.

Finally although cluster analysis sorted accessions into clusters, it is understood that clusters may be biologically meaningless (Edwards & Cavalli-Sforza, 1965). PCA assessed the characters which contributed most to each component. PCA than cluster analysis gave a better picture of the spatial relationships between accessions and was thus useful in complementing and confirming groups obtained through cluster analysis as other authors have found (Jackson & Crovello, 1971; Crossa *et al.*, 1995; Hintum, 1995). It was advantageous therefore to use the two techniques to produce reliable and explainable results.

For further evaluation of the characters which were found useful in the study and the clusters of accessions produced in this study, a larger and more representative sample of the East African Highland bananas must be analysed. This constitutes the next phase of this study.

Chapter 5

MULTIVARIATE ANALYSES OF ACCESSIONS IN THE TWO UGANDA NATIONAL BANANA GERMPLASM COLLECTIONS

5.1 Introduction

The multivariate analyses of morphological variation among the East African Highland bananas in the Kawanda collection reported in the previous chapter have suggested that it is possible to classify the Highland bananas into identifiable clone sets. The Kawanda collection, by the time this study began, was relatively small and contained relatively few accessions from some of the clone sets. It is appropriate therefore to test the applicability of these clone sets with a larger sample of East African Highland bananas and also to attempt to define and delimit the clone sets more precisely using discriminant analysis.

5.2 Objectives

Multivariate methods were therefore applied to a larger sample of accessions of East African Highland bananas by adding data from the Kabanyolo collection to those from Kawanda. The objectives of this phase were 1) to assess whether phenotypic variation prevents accurate classification of East African Highland bananas; 2) to assess and test the applicability of clone sets from the previous study and to define them more precisely.

5.3 Materials and methods

192 accessions from the national banana collections were used in this study. They included the 59 accessions previously used from the Kawanda banana collection and 133 from the Kabanyolo banana collection. The numbers of accessions from each clone set in each collection are shown in Table 5.1. 36 of the accessions belonged to Musakala clone set, 37 to Nakitembe clone set, 65 to Nfuuka clone set, 19 to Nakabululu clone set and 35 to Beer clone set of the subjective classification.

15 pairs of the accessions included in the study were known to have originated as ramets from a single individual. These are listed in Table 3.1, chapter 3. A further 22

pairs of accessions were suspected to be duplicates because they had the same name in both collections. However, these were collected on different occasions and/or from different sites, so were not necessarily true genotypic duplicates.

Five accessions were known to be mutants from accession data records. Three of these were included in the previous study of the Kawanda collection. One additional mutant occurred in the Kabanyolo collection. KM15 (Siira White) had green upper sheaths and midribs whereas the original accession K15 (Siira Red) had deep red coloured upper sheaths and midribs. The fifth mutant KM30 (Enyakinika), was collected from a farmer's field as a mutant of K30 (Nakinyika) but the two looked similar in most aspects during maintenance in the collection.

Four other accessions, K84 and KB84 (Oruhuuna cooking and Oruhuuna beer), and K87 and KB87 (Namunyere cooking and Namunyere beer) were collected as both beer and cooking forms from farmers' gardens. Each beer accession is presumed to be a mutant from the corresponding cooking clone. K84 was subjectively classified as a Nakitembe clone, while K87 was classified as a Nfuuka clone. Their presumed mutants were subjectively classified as Beer clone set. An additional accession, KB140, was said at the time of collection to produce bunches in which some hands were bitter while other hands were not. This accession had no local name so the collectors named it mutant mixed. All hands have been bitter throughout its maintenance in the collection, so it was placed in the Beer clone set of the subjective classification.

Table 5.1: Numbers of accessions in each collection belonging to each of the 5 subjective clone sets.

Subjective clone set	Kawanda collection	Kabanyolo collection	Total
Beer	7	28	35
Musakala	17	19	36
Nakabululu	6	13	19
Nakitembe	12	25	37
Nfuuka	17	48	65
Total	**59**	**133**	**192**

61 characters were used in cluster analysis, compared to the 60 characters of the previous analysis. The colour of fruit skin did not vary among the accessions in Kawanda banana germplasm collection. Since it did vary in this larger sample, it was restored to the analysis. This analysis was therefore based on 13 quantitative and 48 qualitative characters. Of the qualitative characters, 29 were binary, 15 were ordered multistate and 4 were unordered multistate characters.

The pairwise estimation of resemblance between accessions was carried out using the correlation coefficient and the average taxonomic distance coefficient. Cluster analyses of the similarity and dissimilarity matrices were conducted using the group average clustering method since this produced the highest cophenetic values in the previous study. The correlation matrix was used to carry out principal component analysis.

In the discriminant analysis, the same accessions were used but 2 accessions (CB58, KB58) which had missing values were omitted. The first run of the analysis involved a training set of 131 accessions which had been assigned to 5 clone sets (Beer, Musakala, Nakabululu, Nakitembe and Nfuuka) and had been recognised by cluster analysis and the subjective grouping. This training set was used to classify the 59 accessions which were inconsistently placed by cluster analysis and subjective classification. They were treated as a holdout sample.

16 characters (numbers 18, 23, 25, 26, 30, 40, 41, 42, 43, 47, 48, 50, 53, 57, 59, 60) were used in discriminant analysis. 7 characters (numbers 4, 7, 13, 19, 31, 44, 55 in chapter 3, Table 3.3.) which had missing values (one state non-applicable in some accessions) were removed and also all characters that varied within clone sets and distinguished very few accessions. The 16 characters remaining were mainly those which had high loadings on the first component in PCA of the combined data from the two collections and were useful in separating clone sets along that component (see results of PCA).

In discriminant analysis, because the distribution of data within groups was not multivariate normal, a non-parametric k-nearest neighbour method was used to carry out the analysis. Values for k of 5 and 10 were tested to determine which gave the smallest misclassification rate. The effects of varying the number of predetermined classes was also tested. The number of classes varied from 5 (as in the subjective classification) to 6, 7 or 8, if small but apparently distinct groups of accessions were treated as clone sets. These three small clone sets were Siira, Namwezi and Ebihuuna.

Six accessions (K9, C15, K15, KM15, C16 and K90) were treated as the Siira clone set; another 6 accessions (C32, K32, K33, C40, K153 and K154) as Namwezi clone set; and 7 accessions (CM4, C18, K18, K82, K83, K84, KB84) as the Ebihuuna clone set.

5.4. Results

5.4.1 Cluster analysis

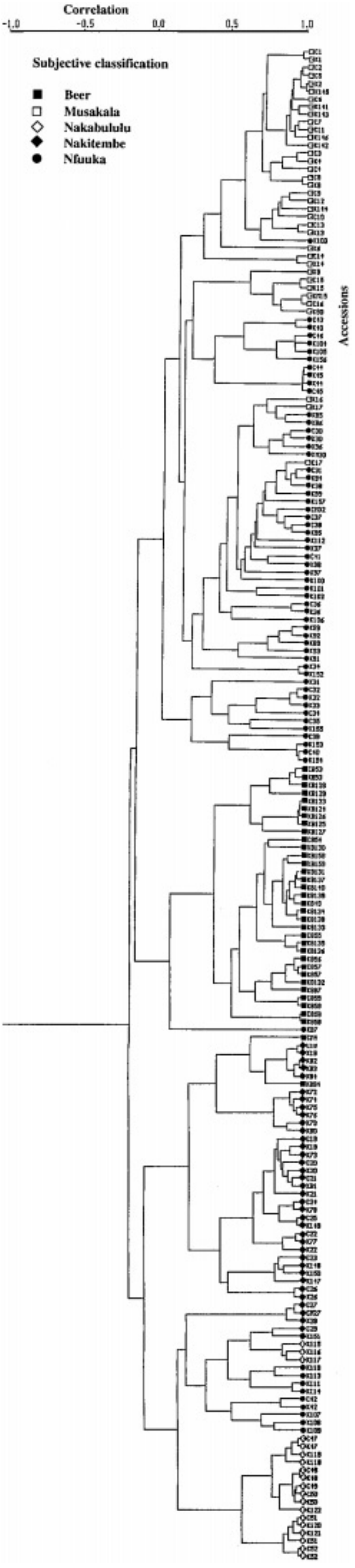
Figures 5.1 and 5.2 represent the phenetic classifications derived from group average clustering of the matrices of correlation and distance coefficients respectively. The two phenograms showed that the accessions in the analyses were not grouped according to the different locations where they were grown. This means that the differences in environments and maintenance practices between Kawanda and Kabanyolo did not cause accessions grown at each site to cluster each according to its location. Instead the accessions clustered in groups similar to those of the subjective classification. 19 of 37 pairs of accessions known or suspected to be duplicates clustered at the first fusions of accessions in both figures 5.1 and 5.2. This indicates that one member of each pair was more similar to its counterpart in the other collection than to any other accession in the analysis confirming further that the differences in growing conditions did not prevent probable duplicates from clustering together.

5.4.2 The validity of the subjective classification

As the number of accessions increased from the 59 which were used in the previous analysis to the 192 used in this analysis, there were two principal outcomes. Firstly, there was an increase in the size and heterogeneity of the 5 clusters corresponding to the subjective clone sets (Figs. 5.1, 5.2). Secondly some accessions which had been outliers in the previous study combined with other accessions in the Kabanyolo collection to which they were phenetically similar, to form distinct clusters which needed consideration as possible clone sets.

Cluster analysis generally agreed with the subjective classification since the majority of accessions assigned to the different subjective clone sets formed distinct clusters in both phenograms (Figs. 5.1, 5.2). However, when the clusters were studied more critically, the position of several accessions did not conform to their subjective classification.

Figure 5.1: Phenogram showing group average cluster analysis of matrix of correlation coefficients between 192 accessions in the Kawanda and Kabanyolo banana germplasm collections (C=accessions from Kawanda; K=accessions from Kabanyolo).



5.4.2.1 Beer clone set

35 accessions were subjectively classified as members of the Beer clone set. 33 of these accessions formed a distinct subcluster in both phenograms (Figs. 5.1, 5.2). These 33 constituted the revised Beer clone set (Table 5.2).

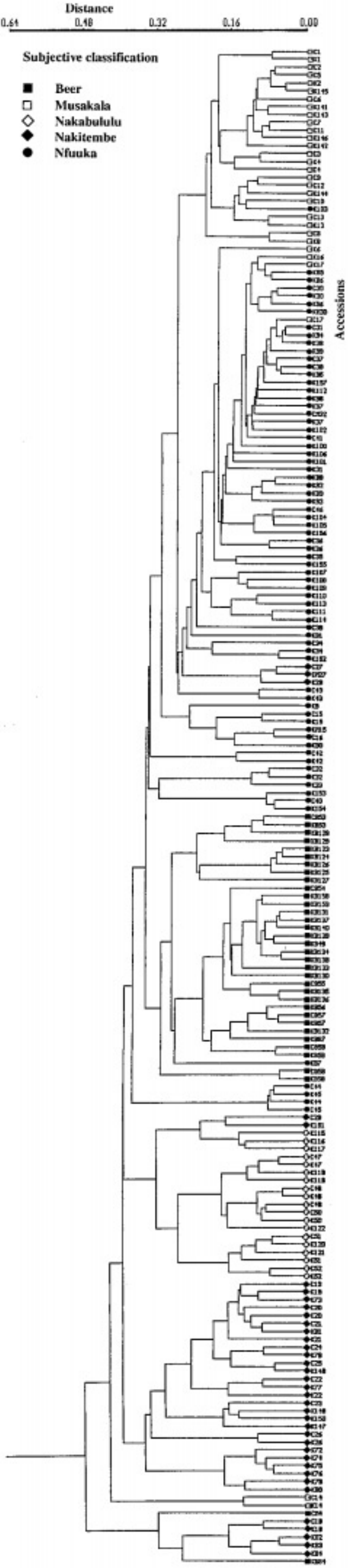
The two beer accessions omitted from this subcluster were CM4 and KB84. As in the previous analysis CM4 (a reported mutant of C4 in Musakala group) did not cluster with the Beer accessions. In both phenograms, CM4 appeared in the same subcluster as another Beer accession, KB84, considered by the farmer from whom it was collected to have arisen from a cooking banana subjectively classified in the Nakitembe group. CM4 and KB84 clustered with other Nakitembe accessions(C18, K18, K82, K83, K84). Together these 7 accessions are considered possible candidates for a new clone set to be called Ebihuuna.

Table 5.2: Numbers of accessions assigned to the various subjective clone sets in the phenograms resulting from group average clustering of the correlation and distance matrices.

Clone set	Subjective classif.	Correlation phenogram	Distance phenogram	Accessions placed in this clone set in all 3 classifications	
				Accession numbers	Total
Beer	35	33	33	KB49, CB53, KB53, CB54, CB55, KB56, CB57, KB57, CB58, KB58, CB59, KB59, KB87, KB123, KB124, KB125, KB126, KB127, KB128, KB129, KB130, KB131, KB132, KB133, KB134, KB135, KB136, KB137, KB138, KB139, KB140, KB158, KB159	33
Musakala	36	27	24	C1, K1, C2, K2, C3, C4, K4, C5, C6, C7, C8, K8, C9, C10, C11, C12, C13, K13, K141, K142, K143, K144, K145, K146	24
Nakabululu	19	16	19	C47, K47, C48, K48, C49, C50, K50, C51, K51, C52, K52, K118, K119, K120, K121, K122	16
Nakitembe	37	32	27	C19, K19, C20, K20, C21, K21, C22, K22, C23, C24, C25, C26, K26, K72, K73, K74, K75, K76, K77, K78, K79, K80, K81, K147, K148, K149, K150	27
Nfuuka	65	43	51	C30, K30, KM30, C31, CM32, K34, C36, K36, C37, K37, C38, K38, C41, K85, K86, K88, K89, K91, K92, K93, K94, K95, K96, K97, K98, K99, K100, K101, K102, K106, K112, K152, K157	33

Accession K87 was an outlier to the beer cluster in both phenetic classifications although it was subjectively classified as a Nfuuka accession. Based on the correlation matrix, K87 was most similar to KB132, a beer clone. According to the distance matrix K87 was most similar to K96 (Nfuuka group). KB87 is a supposed mutant of K87 but it was not most similar to its presumed parent. KB132, the

Figure 5.2: Phenogram showing group average cluster analysis of matrix of distance coefficients between 192 accessions in the Kawanda and Kabanyolo banana germplasm collections (C=accessions from Kawanda; K=accessions from Kabanyolo).



accession to which K87 was most similar, is phenetically very similar to KB87. Their differences occur in the flower structure where the style of KB132 is somewhat hooked and the male inflorescence is pendulous. The style of KB87 is straight and the male inflorescence is more oblique than pendulous. However, as mutants evolve, the parent-mutant relationship gradually becomes less obvious, which may explain K87 not being most similar to its parent.

5.4.2.2 Musakala clone set

Addition of the Kabanyolo accessions increased the number of accessions subjectively classified as Musakala from 17 to 36. There was agreement between the subjective and the 2 phenetic classifications regarding 24 of these accessions (Table 5.2). Both phenetic classifications (Figs. 5.1, 5.2) contained a cluster corresponding to a large part of the subjective clone set Musakala but including K103, which was subjectively classified as Nfuuka. The highest pairwise correlation of K103 was with C10, which is a Musakala accession, and K103 was also least distant from C10. In the study of the Kawanda collection, C8 was either positioned at the boundary of the Musakala cluster or was intermediate between Musakala and Nfuuka. In this analysis, C8 and K8 clustered with Musakala accessions in both phenetic classifications. 3 accessions (K16, K17, C17) subjectively classified as Musakala were included in the Nfuuka cluster in both phenograms. 6 others (K9, C15, K15, KM15, C16 and K90) formed a distinct subcluster which joined the Nfuuka cluster before the Nfuuka and Musakala clusters fused in the correlation phenogram (Fig. 5.1) but which clustered with the Musakala-Nfuuka clone sets after the two clusters had fused in the distance phenogram (Fig.5.2). This is the Siira subcluster, which is an additional new clone set, like Ebihuuna (section 5.4.2.1).

Two other accessions (C14, K14) clustered with Musakala in the phenogram based on the correlation matrix (Fig. 5.1) but were outliers to all groups in the phenogram based on the distance matrix (Fig.5.2). This pattern was the same as that shown by C14 in the previous analysis. Similarly, K6 joined the Musakala cluster in the correlation phenogram but was an outlier to part of the Nfuuka clone set in the distance phenogram.

This analysis has confirmed the finding from the previous analysis that some accessions subjectively classified as Musakala are phenetically more similar to

Nfuuka than to Musakala. These accessions are K9, C15, K15, KM15, C16, K16, K17 and K90. One accession (K103), subjectively classified as Nfuuka, was more similar to Musakala than Nfuuka.

These results suggest that Musakala is more similar to Nfuuka than to the other subjective clone sets. They further suggest that K16, K17 and C17 should be reclassified in the Nfuuka clone set, and that K103 should be reclassified in the Musakala clone set. K6 and K9, C15, K15, KM15 C16 and K90 are intermediate between Nfuuka and Musakala. However, K9, C15, K15, C16 and K90 consistently clustered together and they could form a new clone set, Siira.

5.4.2.3 Nakabululu clone set

Addition of the Kabanyolo accessions increased the number of accessions subjectively classified as Nakabululu from 6 to 19. 16 accessions appeared as a distinct cluster in both phenograms and hence constitute the Nakabululu clone set (Figs 5.1, 5.2; Table 5.2). In both phenograms, 3 accessions subjectively placed in

Nakabululu (K115, K116, K117) clustered with two Nakitembe accessions (C29 and K151), forming a subcluster of their own.

Originally accessions K115, K116 and K117 together with C29 and K151 were subjectively classified as Kibuzi, meaning fat goat. They all had very compact bunches with inflated fruits which had blunt apices. They shared characters with both Nakabululu and Nfuuka accessions. K115, K116 and K117 were later subjectively classified as Nakabululu accessions while C29 and K151 were classified as Nakitembe accessions because they had partially persistent floral parts on their inflorescence rachis.

5.4.2.4 Nakitembe clone set

A total of 37 accessions in this analysis were subjectively classified in the Nakitembe clone set. The subjective and the 2 phenetic classifications agreed regarding 27 accessions, which constitute the provisional Nakitembe clone set (Table 5.2).

In the previous study C26 (subjectively classified as Nakitembe) was either positioned at the boundary of the Nakabululu cluster (Fig. 4.7) or was an outlier to all groups (Fig. 4.8). In this study C26 appeared in the Nakitembe cluster in both phenograms, in agreement with its subjective classification.

C27, CM27 and C29 did not consistently join any other cluster in the previous study. They joined Nfuuka in some of the distance phenograms, while C27 and CM27 joined Nakabululu in one correlation phenogram. In this analysis C27, CM27 and K28 clustered with Nfuuka in the distance phenogram. In the correlation phenogram, they were outliers to the Kibuzi subcluster. C29 and K151 became part of Nakabululu cluster in both phenograms. K151 is a Kabanyolo accession very similar to C29 but with minor differences. It seems to be a dwarf type of C29. Accessions C18, K18, K82, K83, K84 and the two beer bananas, KB84 and CM4 formed their own subcluster. This subcluster linked with the Nakitembe cluster in the correlation phenogram while in the distance phenogram, it was an outlying subcluster. These accessions may constitute a separate clone set, Ebihuuna.

5.4.2.5 Nfuuka clone set

65 accessions were subjectively classified as Nfuuka. The subjective and the two phenetic classifications agreed regarding 33 of these accessions (Table 5.2). The remaining accessions occupied different positions in the two phenograms.

Table 5.3 compares the positions of the inconsistently placed accessions in each phenogram. This Table further shows whether the inconsistently placed accessions are most similar to an accession in the subcluster which they join or to an accession in a different subcluster. The inconsistently placed accessions of Nfuuka clone set were of three types. The first type consisted of single accessions (K31, C35, C39) which changed positions according to the coefficient used to measure similarity (Table 5.3). The second type of accession had the same nearest neighbour as assessed by both distance and correlation coefficients but they were inconsistently placed as a consequence of group average clustering. The majority of clone set accessions in Table 5.3 are of this type e.g C44, K44, C45 and K45. Accessions of the third type were placed in a different cluster from that indicated by their subjective classification in both phenograms. K103 clustered with Musakala accessions and its nearest neighbour was C10 in the Musakala cluster.

Table 5.3: Comparison of positions of Nfuuka accessions which have been inconsistently placed

Accessions	Subcluster in correlation phenogram	Greatest similarity	Subcluster in distance phenogram	Least dissimilarity
K31	with Namwezi subcluster	C35	chained in Nfuuka cluster	C37
C34	with Namwezi subcluster	C32	chained in Nfuuka cluster with K34, K152	C32
C35, K155	with Namwezi subcluster	K155 (for C35) C35 (for K155)	chained in Nfuuka	K155 (for C35) K35 (for K155)
C39	with C40, K153, K154	K153	chained in Nfuuka cluster	K38
C46, K104,	with C43, K43	K105 (for C46 and K104)	chained in Nfuuka cluster	K156(for C46)
K105, K156	then with C44 etc.	C104 (for K105) K156 (for C46) C46 (for K104)		K105 (for K104) C46 (for K104)
K104(C46,K105)				
K107, K108,	with C42, K42+	K107 (for K108)		K108/K109
K109	K110, K111, K113, K114+	K109 (for K107)	with K110, K111, K113	K107/K109
	aberrant Nakabululu and Nakitembe	K109 (for K108)	K114 chained in Nfuuka before junction with Namwezi	K109/K108
K110, K111, K113, K114	C29, K151, K115, K116, K117, C27, CM27, K28	K113 (for K110) K114 (for K111) K111 (for K114) K110 (for K113)	K107, K108 K109 chained in Nfuuka before junction with Namwezi	K111/K114 K114/K111 K113/K110 K110/K113
C32, K32, K33, C40, K154, K153	named Namwezi but formed a larger group with C34, C35, K155, C39	C32(for K32) K33(for K32) C40 (for K154) K153(for K154)	distinct subcluster named Namwezi, joins Musakala/Nfuuka cluster C32, K32, K33	C32 (for K32) K33 (for K32) C40 (for K154) K153 (for K154) K154 (for K153)
C42,K42	K107, K108, K109	K42/C42	C42, K42	C42/K42
C43,K43	with C44, K44, C45, K45, K104, K105, K156	C43/K43	C43, K43	C43/K43
C44, K44, C45, K45	with C43/K43, C44 etc. as separate cluster, very closely related	C44/K45	joins Nfuuka/Beer/ Musakala cluster	C44/K45
K87	with Beer	KB132	with Beer	K96
K103	with Musakala	C10	with Musakala	C10

K87 clustered with Beer accessions in both phenograms but the nearest neighbour of K87 as measured by the distance coefficient is K96 in the Nfuuka cluster.

5.4.2.6 Preliminary discussion

The results from cluster analysis agreed with the subjective classification with regard to the positions of the majority of the accessions in the enlarged sample of the East African Highland bananas. However, 59 of the accessions may need a reclassification to new clone sets or a redefinition of the subjective clone sets (Table 5.4) would be necessary. Further discrimination of clone sets is considered in the results of discriminant analysis. The 5 subjective clone sets proposed are valid and should be retained.

5.4.3 Principal component analysis

Figures of scatter plots from PCA results have not been presented because as the number of accessions increased, groups of accessions became less defined due to much overlapping. However, a brief comment on the results has been made.

Results from PCA indicated that as the number of accessions increased, the amount of variation accounted for by each component became smaller. Component one accounted for 11% of the total variation. It separated the Nakabululu clone set, the combined Beer-Nfuuka clone sets and the Musakala clone set. Component two accounted for only 8% of the total variation responsible for separating Nakabululu clone set further from the rest, the Nakitembe clone set, two beer accessions CM4 and KB84, and C18, C82, C83, C84 from all others in the analysis. Component three accounted for for only 7% of the total variation. It was responsible for separating the Nakabululu clone set further from the rest.

Table 5.4: Accessions from each subjective clone set which might need reclassification as suggested by cluster analyses.

Accessions	Subjective clone set	Suggested reclassification
CM4, KB84	Beer	Ebihuuna clone set
K6, K9, C15, K15, KM15, C16, K90	Musakala	Siira clone set
C14, K14	Musakala	Refer to final classification (Table 5.8)
K16, C17, K17	Musakala	Nfuuka clone set
K115, K116, K117	Nakabululu	Refer to final classification (Table 5.8)
C18, K18, K82, K83, K84	Nakitembe	Ebihuuna clone set
C27, CM27, K28, C29, K151	Nakitembe	Refer to final classification (Table 5.8)
C32, K32, K33, C40, K153, K154	Nfuuka	Namwezi clone set
K31, C34, C35, K155	Nfuuka	Refer to final classification (Table 5.8)
K103	Nfuuka	Musakala clone set
C39	Nfuuka	Refer to final classification (Table 5.8)
C42, K42	Nfuuka	“
C43, K43	Nfuuka	“
C44, K44, C45, K45	Nfuuka	“
C46, K104, K105, K156	Nfuuka	“
K107, K108, K109	Nfuuka	“
K110, K111, K113, K114	Nfuuka	“

On the whole, the clone sets were not well defined as there was more overlapping of clone sets than in the previous study. PCA however provided useful characters for separating clone sets (Table 5.5).

Of the 61 characters used in the study, 10 characters accounted for much of the variation on component one. 5 of these were used also in the subjective classification (Table 5.5). The characters which had positive loadings on component one were useful in separating Musakala clone set. These included bunch orientation and shape, fruit

apices, fruit length/width ratio, fruit shape and rachis position. These were the same characters that separated Musakala clone set in the previous study. However, twisting of the leaf tip did not contribute as much as it did in the previous study.

Three other characters with negative loadings above 0.5 were important in separating Nakabululu group along the first component. These were bunch compactness, fruit position in a bunch and fruit cracking. The first two were important in the previous study as well. Bunch compactness was found to be a useful character in defining Nakabululu clone set.

Table 5.5: Characters with loadings greater than 0.5 (in bold) on principal components 1, 2, 3 or 4.

Character	PC 1	PC 2	PC 3	PC4
Undersheath colour	-0.034	0.063	-0.207	**0.654**
Pseudostem blotches	-0.010	0.001	-0.096	**0.783**
*Bunch orientation	**0.855**	0.043	-0.066	0.049
Bunch shape	**0.718**	-0.023	-0.275	0.081
*Bunch compactness	**-0.866**	0.058	0.079	-0.049
Fruit position in a bunch	**-0.812**	0.037	-0.200	-0.037
*Pulp colour at maturity	-0.236	**-0.722**	0.118	-0.345
*Fruit apex	**0.744**	-0.068	-0.173	-0.020
Style on fruit apex	-0.100	**-0.757**	0.074	0.157
Type of persistent style	0.004	0.065	**-0.960**	-0.067
Persistent stamen	-0.104	**-0.560**	-0.457	0.120
Fruit cracking	**-0.661**	-0.012	0.397	-0.020
*Fruit length/width ratio	**0.687**	0.034	-0.057	-0.088
Fruit shape	**0.814**	-0.104	-0.320	0.013
*Inflorescence rachis position	**0.718**	-0.060	-0.435	0.071
*Persistent neuter flowers	0.049	**-0.861**	0.078	0.164
Male bud anthocyanin	-0.319	**-0.523**	-0.146	-0.055
*Male bud shape	-0.440	-0.372	**-0.585**	0.028
Male bud waxiness	-0.278	**0.649**	-0.020	0.069
Male bud apex	**-0.504**	-0.346	**-0.575**	0.006
*Bract imbrication	0.116	**-0.758**	-0.205	0.152

*** characters used in subjective classification**

Seven characters with loadings above 0.5 were important on component two. Five of these with negative loadings were responsible for separating the Nakitembe clone set. These included pulp colour at maturity, persistent style and stamens on fruit apices, persistent neuter flowers along the rachis and bract imbrication. Male bud anthocyanin and waxiness were important characters in separating two Beer accessions (CM4 and CB84) and five Nakitembe accessions (C18, K18, K82, K83, K84) on component two.

Three characters had loadings above 0.5 along this component three. These included type of persistent style, which separated some Nakitembe accessions (C18, K18, K82, K83,K84) and two Beer accessions CM4 and CB84. The male bud shape and apex contributed to the separation of Nakabululu clone set. Much of the variation on component four was contributed by two characters. These had loadings above 0.5 and were the background appearance of the pseudostems and the presence or absence of blotches. The two characters were responsible for separating C32, K32 and K33 from the rest of the accessions.

5.4.4 Discriminant analysis

Results of cluster analysis have shown that 133 of the 192 accessions under study have been phenetically assigned to the five clone sets of the original subjective classification. 59 accessions (Table 5.4), however, occupied different positions in the subjective classification and the two phenetic classifications. Discriminant analysis was therefore used to classify these 59 accessions into predetermined clone sets. 59 accessions represented the holdout sample and 133 accessions represented the training set.

In successive runs of this analysis with different numbers of clone sets and different values of k, there were several outcomes (Table 5.6). Ebihuuna, Namwezi and Siira were small clone sets (6-7 accessions). With k=10, each had 3-4 nearest neighbours in another clone set and the percentage misclassification rate was thus higher than when k=5 (Table 5.6).

With k=5 the misclassification rates varied with the number of clone sets recognised. The lowest misclassification rates were obtained with 5 clone sets and with the 6 clone sets with Ebihuuna being the sixth. When accessions from the farmers' fields were added to the analysis, there was no misclassification with 5 clone sets but a misclassification rate of 0.0046 for the 6 clone sets. The classification into five clone sets was therefore retained. All the accessions in the training set were correctly classified, agreeing with results from cluster analysis and the subjective classification.

Table 5.6: Misclassification rates at different values of k and different numbers of clone sets.

Number of clone sets	Misclassification rates with different values of k (upper figure in each cell represents stratified error rate for resubstitution and lower figure represents stratified error rate for crossvalidation)	
	5	10
5 clone sets (as in original subjective classification)	0.0025 0.0031	0.0031 0.0025
6 clone sets 5+Ebihuuna	0.0029 0.0026	0.0228 0.0232
6 clone sets 5+Siira	0.0088 0.0140	0.0333 0.0415
6 clone sets 5+Namwezi	0.0569 0.0625	0.0738 0.0806
7 clone sets 5+Ebihuuna+Siira	0.0083 0.0134	0.0496 0.0571
7 clone sets 5+Ebihuuna+Namwezi	0.0542 0.0594	0.0879 0.0944
7 clone sets 5+Namwezi+Siira	0.0560 0.0662	0.0941 0.1081
8 clone sets 5+Ebihuuna+Namwezi+Siira	0.0534 0.0631	0.1067 0.1201

5.4.4.1 Distances between clone sets

Table 5.7 shows squared distances between clone sets. The distances between clone sets were all significantly different from distances within clone sets. Nfuuka and Nakabululu were the least distant clone sets, followed by Nfuuka and Nakitembe. Beer was the most distant. Results from cluster analysis suggested that Musakala and Nfuuka were phenetically closer to each other than either was to any other clone set. This was indicated by the number of accessions which changed positions between the two major

clusters of Nfuuka and Musakala or occupied boundaries of the main Nfuuka and Musakala clusters. However, one disadvantage of cluster analysis is that it cannot show clearly the distances between clone sets, only between the most similar ones. One accession or clone set can cluster with only one accession or clone set at a time.

5.4.4.2 Classification of unclassified accessions

Table 5.8 gives the results of the classification of the unclassified accessions in the holdout sample and the posterior probability of each accession belonging to the clone set in which it has been placed. The posterior probabilities were 1.0 for each accession except for C29 which had posterior probability of 0.6 of belonging to Nakabululu and a posterior probability of 0.3 of belonging to Nfuuka.

Table 5.8 also shows both the subjective and phenetic classifications of these accessions. Ten of the 12 accessions in the holdout sample subjectively classified as Musakala were classified as Nfuuka accessions by the discriminant analysis. The remaining two (C14, K14) remained in Musakala. Five accessions (C27, CM27, K28, C29 and K151 subjectively classified as Nakitembe, were reclassified as Nakabululu accessions. Accessions which had been regarded to be distinct as Ebihuuna, two had been subjectively classified as Beer and were retained as Beer accessions by discriminant analysis while the five remained as Nakitembe accessions as well. The accessions which were regarded as Namwezi were also retained as Nfuuka accessions. However, the accessions which were regarded as Siira clone set had been subjectively classified as Musakala and discriminant analysis placed them under the Nfuuka clone set.

Table 5.7: Squared distances between the five clone sets. All the F-statistics are significant at P<0.0001.

Clone sets	Beer	Musakala	Nakabululu	Nakitembe	Nfuuka
Beer	-				
Musakala	4447	-			
Nakabululu	2773	1674	-		
Nakitembe	2772	1674	445.1	-	
Nfuuka	2773	1674	44.7	383.6	-

Table 5.8: The classification of the 53 accessions in the holdout sample and the posterior probabilities that they have been correctly classified by classificatory discriminant analysis.

Accession	Subjective classification	Cluster analyses	Classificatory discriminant analysis	Posterior probability
CM4	Beer (Ebihuuna)	inconsistent position	Beer	1.0
KB84	Beer (Ebihuuna)	“	Beer	1.0
K6	Musakala	“	Nfuuka	1.0
K9	Musakala (Siira)	“	Nfuuka	1.0
C14	Musakala	“	Musakala	1.0
K14	Musakala	“	Musakala	1.0
C15	Musakala (Siira)	“	Nfuuka	1.0
K15	Musakala (Siira)	“	Nfuuka	1.0
KM15	Musakala (Siira)	“	Nfuuka	1.0
C16	Musakala (Siira)	“	Nfuuka	1.0
K16	Musakala	Nfuuka	Nfuuka	1.0
C17	Musakala	“	Nfuuka	1.0

Table 5.8 (cont'd.)

Accession	Subjective classification	Cluster analyses	Classificatory discriminant analysis	Posterior probability
K17	Musakala	"	Nfuuka	1.0
K90	Musakala (Siira)	"	Nfuuka	1.0
K115	Nakabululu	inconsistent position	Nfuuka	1.0
K116	Nakabululu	"	Nfuuka	1.0
K117	Nakabululu	"	Nfuuka	1.0
C18	Nakitembe (Ebihuuna)	"	Nakitembe	1.0
K18	Nakitembe (Ebihuuna)	"	Nakitembe	1.0
C27	Nakitembe	"	Nakabululu	1.0
CM27	Nakitembe	"	Nakabululu	1.0
K28	Nakitembe	"	Nakabululu	1.0
C29	Nakitembe	inconsistent position	Nfuuka Nakabululu	0.33 0.66
K82	Nakitembe (Ebihuuna)		Nakitembe	1.0
K83	Nakitembe (Ebihuuna)	"	Nakitembe	1.0
K84	Nakitembe (Ebihuuna)	"	Nakitembe	1.0
K151	Nakitembe	"	Nfuuka	1.0
K31	Nfuuka	"	Nfuuka	1.0
C32	Nfuuka (Namwezi)	"	Nfuuka	1.0
K32	Nfuuka (Namwezi)	"	Nfuuka	1.0
K33	Nfuuka (Namwezi)	"	Nfuuka	1.0
C34	Nfuuka	"	Nfuuka	1.0
C35	Nfuuka	"	Nfuuka	1.0
C39	Nfuuka	"	Nfuuka	1.0
C40	Nfuuka (Namwezi)	"	Nfuuka	1.0
C42	Nfuuka	"	Nfuuka	1.0
K42	Nfuuka	"	Nfuuka	1.0
C43	Nfuuka	"	Nfuuka	1.0
K43	Nfuuka	"	Nfuuka	1.0
C44	Nfuuka	"	Nfuuka	1.0
K44	Nfuuka	"	Nfuuka	1.0
C45	Nfuuka	"	Nfuuka	1.0
K45	Nfuuka	"	Nfuuka	1.0
C46	Nfuuka	"	Nfuuka	1.0
K87	Nfuuka	Beer	Nfuuka	1.0
K103	Nfuuka	Musakala	Nfuuka	1.0
K104	Nfuuka	inconsistent position	Nfuuka	1.0
K105	Nfuuka	"	Nfuuka	1.0
K107	Nfuuka	"	Nfuuka	1.0
K108	Nfuuka	"	Nfuuka	1.0
K109	Nfuuka	"	Nfuuka	1.0
K110	Nfuuka	"	Nfuuka	1.0
K111	Nfuuka	"	Nfuuka	1.0
K113	Nfuuka	"	Nfuuka	1.0
K114	Nfuuka	"	Nfuuka	1.0
K153	Nfuuka (Namwezi)	"	Nfuuka	1.0
K154	Nfuuka (Namwezi)	"	Nfuuka	1.0
K155	Nfuuka	"	Nfuuka	1.0
K156	Nfuuka	"	Nfuuka	1.0

5.4.5 Discussion

One of the objectives of this phase was to assess whether phenotypic variation would prevent accurate classification of the East African Highland bananas. Results from cluster analysis have shown that different management conditions at the two locations where the accessions were grown did not cause accessions grown at the same location to cluster together. Known ramets clustered together even though they were maintained in different collections. Three-quarters of the characters were qualitative and one-quarter were quantitative. Of these two types of characters, 21 had a loading on either the first, second, third or fourth principal component of more than 0.5 (Table 5.5). Only one quantitative character (fruit length/width ratio) contributed significantly to the separation of accessions into groups. Quantitative characters are much more affected by the environment than qualitative ones (Crane & Lawrence, 1956; Bell, 1967; Ayala, 1982; van Hintum, 1995).

Although the growing conditions did not prevent the classification of the Highland bananas in previously identified clone sets, 59 accessions occupied different positions with regard to the subjective classification and cluster analysis. A majority of these accessions occupied positions intermediate between clone sets according to PCA as previously discussed in chapter four. This indicated that they had some of the characters of different clone sets and hence cannot directly cluster with any.

While cluster analysis can identify clusters of phenetically related accessions, it fails to delimit some in a precise manner. Intermediate accessions join any one of the clone sets it shares characters with but not all. PCA places them somewhere in between groups to indicate what position they specifically occupy in relation to all other accessions.

Classificatory discriminant analysis assigned all accessions to clone sets in a satisfactory manner. Of 190 accessions in the analysis, 189 (99%) were classified correctly (i.e were not misclassified). One accession, C29, which had some characters of Nakabululu and some Nfuuka characters had 66% probability of belonging to Nakabululu and a 33% probability of belonging to Nfuuka.

Classificatory discriminant analysis provides a sorting procedure among accessions to enable groups to be defined more precisely. This is what it exactly did.

The method defined positions of each accession in a particular clone set and provided percentage probabilities of each accession to belong to that clone set.

This is the advantage of the method but we should not forget that it needs some groups to start with, which have been generated by cluster analysis.

Cluster analysis suggested that Musakala and Nfuuka were closer to each other than to other clone sets (Table 5.7). Discriminant analysis showed that Musakala and Nfuuka were not the closest clone sets. Nfuuka and Nakabululu were the closest, followed by Nfuuka and Nakitembe. Beer and Musakala were distant from the other three clone sets. Cluster analysis identifies groups but they may not necessarily be natural groups.

Looking at the way accessions were classified, a brief comment is made. Two beer accessions (CM4 and KB84) were associated with Ebihuuna, a subcluster of Nakitembe, in cluster analysis. The two were classified as Beer accessions by discriminant analysis. Beer clones are separated by the astringency and bitterness of fruits from the cooking clones (Chapter 1, Sebasigari, 1987). KB84 was reported to be a beer mutant of K84, another Ebihuuna accession, by the farmer from whom it was collected and the two look morphologically alike. CM4 was associated with the Nakitembe (Ebihuuna) accessions C18 and K18 in cluster analyses. It has been argued by some farmers that Beer clones

are not mutants of cooking clones and that farmers' worries that their cooking clones are changing to Beer clones are not justified. Beer clones could be mutants which diversified a long time ago from the cooking clones and thus very different from their probable parental accessions in other clone sets, as demonstrated by the large distances between the Beer clone set and other clone sets (Table 5.8). In that sense farmers are right since they never knew Beer clones could have evolved from the cooking clone and these results support them. However, the association of CM4 with C18 and K18 as well as KB84 and K84 in cluster analysis and PCA suggest that CM4 and KB84 could be recent diversification from the cooking bananas. It would be interesting to look at other differences between the C18/K18 and CM4 as well as K84 and KB84, which are not based on morphology.

Musakala is a clone set which is also very different from other clone sets and hence was also at a distance from the rest. Accessions of Musakala have giant bunches which are very lax with the longest and largest fruits among the 5 clone sets. They have been selected through time. Diversification among the Highland bananas still continues. Ebihuuna, Namwezi, Siira and the C14/K14 are other potential future clone sets. The accessions which had been subjectively classified as Musakala were transfered to Nfuuka by discriminant analysis but had one or two characters of Musakala although not necessarily lax bunches with long fruits.

The majority of the accessions unclassified after cluster analysis were classified as Nfuuka accessions by classificatory discriminant analysis. Nfuuka is a controversial clone set. In cluster analysis it was the most diverse clone set with some of its accessions scattered throughout both phenograms forming subclusters (Figs. 5.1, 5.2). In PCA it occupied the central position with respect to the other clone sets. Nfuuka is widely distributed clone set, hence may be the most diverse. It would be interesting to evaluate this clone set further using molecular studies and investigate the possibilities of this clone set being responsible for the generation of other clone sets through mutation and thus having been the earliest clone set.

Nakabululu and Nakitembe were not far from each other and from Nfuuka. Farmers generally believe that Nakitembe and Nakabululu could be the oldest East African Highland bananas. In both cluster analysis and PCA Nakabululu was the most distinct clone set. Its members are very well defined and it looked as if it was a clone set which separated from Nfuuka a long time ago. However, it is very difficult to speculate on such an idea without any further advanced studies related to origins of the crop. Farmers' information should compliment results from such findings.

On the whole therefore, the subjective clone sets were found to be valid since they were recognised by all the multivariate methods which were used.

The 59 accessions were difficult to classify precisely either subjectively or by cluster analysis. This is because they shared characters of different clone sets. Methods which use a probabilistic approach to classify individuals are preferred in handling cases known to be intermediate between groups. This includes classificatory discriminant analysis and MUSAID (Perrier & Tezenas, 1988) in which the likelihood of an unclassified accession belonging to any one of the previously recognised clone sets is calculated. Discriminant analysis has therefore an advantage over cluster analysis and PCA in that it classifies accessions a bit more precisely than cluster analysis and principal component analysis. Discriminant analysis needs groups which have already been generated from cluster and principal component analysis.

Principal component analysis has the advantage of showing how accessions and clone sets are phenetically related by indicating how distant each accession is from all others. Principal component analysis also gives the variables most responsible for giving that pattern of relationships among accessions (Table 5.5). Cluster analysis identifies clusters among individuals based on the given variables and this was our first objective. Principal component analysis and discriminant analysis can then support or not support the clusters generated by cluster analysis. Cluster analysis can further indicate how similar each accession is to each other, a procedure found to be very useful in distinguishing accessions likely to be phenetic duplicates or clones in the next chapter. Cluster analysis, principal component analysis and discriminant analysis complement each other and it is therefore useful to apply them simultaneously.

Chapter 6

ESTIMATION OF NUMBER OF DISTINCT CLONES IN THE TWO UGANDA NATIONAL BANANA GERMPLASM COLLECTIONS AND PROPOSED COMPOSITION OF A CORE COLLECTION

6.1 Introduction

The multivariate analyses of morphological variation among the East African Highland bananas in the two national collections reported in the previous chapter have identified various clone sets of accessions within the Highland bananas. Further more, the analysis confirmed that the different growing conditions of the two collections could not prevent the classification of accessions in their appropriate clone sets.

Multivariate methods have also been found useful both in detecting duplicates within a collection (Ramey *et al.*, 1988) and in identifying clones with different names which may be synonyms (Engels, 1986). This can lead to reduction in size of the collections, which is an advantage since evaluation is easier, faster and more effective with a smaller collection than with a bigger collection. Synonymy in the East African Highland bananas has been reported (Tothill, 1940; Shepherd, 1957), hindering classification and identification within the crop because no charts are available that list the synonyms. The need to distinguish distinct clones became apparent when urgent selection of clones to be used in studies related to disease and pest resistance became necessary. This chapter is divided into two parts. The first concerns estimating the number of distinct clones in the national collections and the second contains a proposal for establishing a core collection.

6.2 Estimating the number of clones in the national collections

Multivariate methods were applied to the same sample of accessions used in the previous study. The objectives of this phase were 1) to assess whether multivariate methods can detect duplicates and identify accessions which are phenetically similar but have different names and 2) to determine the amount of duplication and to estimate the number of distinct clones in the collections.

6.3 Materials and methods

192 accessions from the national banana collections used in the previous study were employed in this study.

15 pairs of accessions as previously indicated (chapter 5) were known to have originated as ramets from a single individual. Four pairs were from the Beer clone set, three from Musakala, one from Nakabululu, one from Nakitembe and six from Nfuuka clone set (Table 6.1). A further 22 pairs were suspected to be duplicates because they had the same name in both collections (Table 6.1). These were collected on different occasions and/or from different sites, so were not necessarily true genotypic duplicates.

Table 6.1: Pairs of known ramets and accessions with the same name in each clone set.

Clone set	Number of pairs of ramets	Number of pairs of accessions with the same name
Beer	4	0
Musakala	3	8
Nakabululu	1	4
Nakitembe	1	5
Nfuuka	6	5
Total	**15**	**22**

Five accessions were known to be mutants from accession data records and these were also included in the previous study (Chapter 5).

The pairwise estimation of resemblance between accessions was carried out using the correlation coefficient and the average taxonomic distance coefficient and the two matrices resulting from the above comparisons were the major elements used in this study.

6.4. Results

6.4.1 Accessions known to be ramets of the same clone

The ranges of values for the correlation and distance coefficients between all pairs of known ramets are shown in Table 6.2. Members of 9 of the 15 pairs of ramets were more similar to each other than to any other accession in the analyses. In the other 6 pairs of ramets, C17/K17, C21/K21, C37/K37, C38/K38, C44/K44 and C45/K45, at least one member of each pair resembled other accessions in the analyses more than its counterpart ramet (Table 6.3). Members of these pairs of ramets differed in more than half of the quantitative characters used, but not in any of their qualitative characters (Table 6.4). As indicated earlier, quantitative characters are particularly likely to be affected by changes in growing conditions (De Langhe, 1961; Bell, 1967; van Hintum, 1995).

Table 6.2: Correlation and distance coefficients between members of each pair of known ramets. (*ramets which were more similar to each other than to any other accession in the analysis).

Ramets	Correlation coefficient between members of each pair	Distance coefficient between members of each pair
C8,K8*	0.959	0.080
C13,K13*	0.934	0.082
C17,K17	0.845	0.075
C21,K21	0.855	0.146
C37,K37	0.759	0.095
C38,K38	0.834	0.070
C42,K42*	0.868	0.147
C43,K43*	0.922	0.098
C44,K44	0.973	0.070
C45,K45	0.965	0.081
C48,K48*	0.988	0.044
CB53,KB53*	0.950	0.086
CB57,KB57*	0.975	0.056
CB58,KB58*	0.988	0.052
CB59,KB59*	0.975	0.060
Range	**0.759-0.988**	**0.044-0.147**
Average	**0.918**	**0.083**

Table 6.3: Correlation and distance coefficients between a ramet and those accessions to which it was more similar than it was to its counterpart ramet.

Ramets	Pairwise correlation between ramets	Correlation with other accessions	Pairwise distance between ramets	Distance from other accessions
C21,K21 (Nakitembe)	0.855	C21, C20 = 0.900 C21, K20 = 0.904 C21, K81 = 0.941 K21, K81 = 0.857	0.146	C21, C20 = 0.115 C21, K20 = 0.117 C21, K81 = 0.089 K21, K81 = 0.145
C44,K44 C45,K45 (Nfuuka)	0.973 0.965	C44, K45 = 0.978 K44, K45 = 0.973 C44, C45 = 0.971 K44, C45 = 0.970	0.070 0.081	C44, K45 = 0.063 K44, K45 = 0.070 C44, C45 = 0.073 K44, C45 = 0.074
C17,K17 (Nfuuka)	0.845	C17, C31 = 0.903 C17, K94 = 0.879	0.075	C17, C31 = 0.055 C17, K94 = 0.061
C37,K37 (Nfuuka)	0.759	C37, C38 = 0.899 C37, K38 = 0.861 C37, K95 = 0.822 C37, K98 = 0.767 C37, K99 = 0.798 K37, K38 = 0.773	0.095	C37, C38 = 0.055 C37, K38 = 0.057 K37, K38 = 0.092 C37, K95 = 0.071 C37, K98 = 0.079 C37, K99 = 0.076
C38,K38 (Nfuuka)	0.834	C38, K95 = 0.907 K38, K94 = 0.928 K38, K95 = 0.842	0.070	C38, K95 = 0.054 K38, K94 = 0.045 K38, K95 = 0.067

Table 6.4: Characters differing between known ramets and the accessions to which these were more similar than they were to their counterpart ramets (the first figure in each comparison represents quantitative characters while the second figure represents qualitative characters)

a) Nakitembe group
C21/K21 (intraclone comparison: 10/0)

Ramets	Other similar accessions		
	C20	K20	K81
C21	12/0	12/0	12/0
K21	11/0	13/0	9/0

b) Nfuuka group
i) C44/K44 and C45/K45 (intraclone comparison C44/K44: 12/0, C45/K45: 13/0)

Ramets	Other similar pair of ramets	
	C45	K45
C44	12/0	11/0
K44	12/0	11/0

ii) C17/K17 (intraclone comparison: 8/0)

Ramets	Other similar accessions	
	C31	K94
C17	11/2	10/2
K17	11/2	10/2

iii) C37/K37 (intraclone comparison: 10/0)

Ramets	Other similar accessions				
	C38	K38	K95	K98	K99
C37	9/2	12/2	11/0	12/0	13/0
K37	12/2	11/2	10/0	12/0	13/0

iv) C38/K38 (intraclone comparison: 10/0)

Ramets	Other similar accessions	
	K94	K95
C38	11/2	11/2
K38	12/2	12/2

6.4.1.1 Nakitembe clone set

C21 and K21 were ramets of a single clone in the Nakitembe clone set named Namulondo in the Luganda dialect of the central region of Uganda. C21 was more similar to 3 other accessions in the Nakitembe clone set (C20/K20 and K81) than to its ramet, K21. K21 was also more similar to K81 than to its ramet, C21 (Table 6.3). According to farmers, C21/K21 is generally a weak clone. Even if it is maintained in good conditions, it does not thrive. Given the facts that mulching and pest control were rare in the Kabanyolo collection and K21 was growing in a block in the collection which was badly eroded, K21 was likely to differ from C21. C20/K20 were not known

to be ramets but shared the name Nakitembe-Nakamali, which means small Nakitembe in Luganda dialect. They originated from the central region and were probably small-statured mutants of Nakitembe (C19). C20/K20 were identical with C21/K21 in all their qualitative characters (Table 6.4a) and therefore probably belonged to the same clone. K81 is alleged by farmers in the zones east of Lake Victoria, in Uganda, to be similar to C21/K21 although it has a different name, Nakangu, because the dialect in this part of Uganda is Lusoga. K81 was also identical to K21 and C21 in all the qualitative characters (Table 6.4a) and hence is assumed to belong to the same clone.

6.4.1.2 Nfuuka clone set

For two pairs of ramets C44/K44 and C45/K45, similarities between the pairs were as great as, or greater than, similarities within each pair (Table 6.3). The 4 accessions were identical in all their qualitative characters (Table 6.4b) and therefore represented a single clone. This conclusion was supported by some farmers in the south western highlands where the clone is widely grown. Some farmers call the clone Enzirabahima (C44/K44), others call it Entukura (C45/K45), but they would state that these 2 names mean the same banana clone.

Differences within the pairs C17/K17 and C37/K37 can be explained by differences in the growing conditions. K17 and K37 were growing in the third block of the Kabanyolo collection which had been badly affected by soil erosion and irregular mulching, leading to great loss of moisture and fertility in the soil.

C17 and K17 were very similar to C31 and K94 (Table 6.3). However they differed from C31 and K94 in two qualitative characters. C17/K17 were therefore not the same clone as C31 or K94. Other differences between C17/K17 and K31 and K94 involved the 13 quantitative characters (Table 6.4b.ii).

C37/K37 were very similar to C38/K38 but differed in colour of fruit pulp and therefore were not the same clone as C38/K38. C37/K37 were also very similar to K95, K98 and K99 (Table 6.4b.iii). These accessions did not differ in any of the qualitative characters and were probably members of a single clone. C37/K37 (Enyeru) is one of the widely distributed clones in Uganda (Karamura *et al.*, 1996). It suckers profusely and matures very fast. Because of its wide distribution, it seems to have a wide range of names and it is not surprising that K95 (Likhago) from the east, and K98 (Sitakange) and K99 (Serunjogi) from the central region may represent this same clone. The name Sitakange is commonly applied to a different clone, in the Nakitembe clone set and that is where it first originated. It means “I will not part with what is mine". The Nakitembe clone has persistent stamens and styles to which the name refers. K98 has no persistent neuter flowers along the rachis so K98 could have been mislabelled.

K99 is a clone commonly known to have a bright pseudostem with fewer black blotches than Enyeru or Likhago. But the pseudostem darkens after its first year ratoon cycle. This change has not been efficiently studied. During record taking Serunjogi had a dark pseudostem.

C38 had its greatest similarity with K95 while K38 was more similar to K94 and K95 than to its ramet C38 (Table 6.3). C38 and K38 differed from K94 and K95 in the colour of the fruit pulp both before and after maturity. C38/K38 therefore belonged to a different clone from both K94 and K95. But what could have caused the ramets C38 and K38 to appear different? C38 had replaced a row of plants in the Kawanda collection which had been infected with weevils. For 18 months after planting, plants of

C38 did not thrive. On the other hand, K38 was growing in one of the new blocks in the Kabanyolo collection, where mulching had been partly carried out and the plants in that block were very healthy. There was therefore a marked difference in the growing conditions of C38 and K38 and this could have created differences between the two.

The name Nfuuka was originally given by farmers to a single clone which readily changed due to different environmental conditions. More clones of this type are now known. They have different names across the banana-growing regions. These clones are very similar morphologically but differ in one or two characters like colour of fruit pulp. This is a difference between C37 and C38 and these are the sorts of changes farmers were referring to. The frequency of these changes have been recorded and discussed at the end of the chapter.

6.4.1.3 Comparison of effects of distance versus correlation coefficients

Some pairs of ramets appeared very similar on the basis of their correlation coefficients, but much less similar on the basis of their distance coefficients, while the reverse was true for some other pairs. This was particularly evident in respect of the pairs of ramets subjected to the greatest differences in growing conditions (C21/K21, C17/K17, C37/K37, C38/K38).

For example C21/K21 and C17/K17 had similar values for their pairwise correlation coefficients, but the distance coefficient between C21 and K21 was much larger than that between C17 and K17. C21 differed from K21 in 10 quantitative characters whereas K17 differed from C17 in only 8 quantitative characters (Table 6.4b.ii). Moreover, the sizes of the differences in these characters were larger in the comparison between C21 and K21 than in the comparison between C17 and K17. This means that the two coefficients interpret the same data differently. The distance coefficient emphasizes differences in size more than the correlation coefficient, as has been noted by others (Sneath & Sokal, 1973).

6.4.2 Accessions sharing the same name but not known to be ramets

The results for the known ramets has given an assessment of the levels of dissimilarity which can be found among ramets due to different environmental conditions. These values of dissimilarity will be useful in assessing whether or not pairs of accessions which share the same name are ramets of a single clone.

6.4.2.1 Accessions which probably represent a single clone

Ten of the twenty two pairs of accessions which shared names, and were given the same identifying numbers, were more similar to each other than to any other accession in the analysis. These were C1/K1, C14/K14, C15/K15, C18/K18, C26/K26, C30/K30, C32/K32, C36/K36 C47/K47 and C52/K52 (Table 6.5). Their pairwise correlation and distance coefficients were within the ranges of values of known ramets. Members of each pair differed in most of their quantitative characters but were identical in all the qualitative characters that were assessed, so the members of each pair were considered to be ramets of a single clone.

Seven other pairs (C2/K2, C4/K4, C19/K19, C20/K20, C22/K22, C50/K50, C51/K51) had pairwise correlation and distance coefficients within the range of those of known ramets.

Table 6.5: Correlation and distance coefficients between accessions sharing the same name but not known to be ramets.

Accs.	Pairwise correlation coefficients	Pairwise distance coefficients	Differences in quantitative/ qualitative characters	Conclusion
C1,K1	0.959	0.075	12/0	ramets
C2,K2	0.925	0.093	10/0	ramets
C4,K4	0.814	0.160	11/0	ramets
C6,K6	0.364	0.236	12/3	different clones
C9,K9	0.400	0.279	11/4	different clones
C14,K14	0.929	0.129	11/0	ramets
C15,K15	0.951	0.093	12/0	ramets
C16,K16	0.313	0.239	12/4	different clones
C18,K18	0.973	0.087	12/0	ramets
C19,K19	0.857	0.136	10/0	ramets
C20,K20	0.830	0.154	12/0	ramets
C22,K22	0.866	0.170	11/0	ramets
C26,K26	0.958	0.099	12/0	ramets
C30,K30	0.893	0.062	12/0	ramets
C31,K31	0.208	0.166	12/0	?
C32,K32	0.958	0.076	12/0	ramets
C34,K34	0.666	0.198	11/0	ramets
C36,K36	0.915	0.077	10/0	ramets
C47,K47	0.978	0.055	11/0	ramets
C50,K50	0.949	0.095	9/0	ramets
C51,K51	0.929	0.119	10/0	ramets
C52,K52	0.979	0.072	12/0	ramets
Range for known ramets	**0.759-0.988** **0.044-0.147**			

However, at least one member of each pair was more similar to other accessions in the analysis than to the counterpart with which it shared its name (Table 6.6). This suggests that accessions with different names can also be ramets as well as those which share names. These seven pairs are discussed under the clone sets to which they were subjectively assigned.

6.4.2.1.1 Musakala clone set

C2 and K2 (Musakala from central region) were identical in all qualitative characters but differed in the quantitative characters. Differences between C2 and K2 were likely to have been caused by the growing conditions since K2 was growing in the eroded first block of the Kabanyolo collection. C2 and K2 were also identical in qualitative characters to K145 (Bandangeyo from the south-western region) and C5 (Luwata from the central region) (Table 6.6). C5, however, was reported by farmers to have fruits which were more angular than those of C2/K2. The name Luwata relates to angularity of fruits, which makes peeling easier. In the collection the angularity of fruits was not as pronounced as farmers indicated. C5, K145, C2 and K2 probably represented a single clone. Musakala, Bandangeyo and Luwata are therefore synonyms.

C4 and K4 (Mudwale) represent a commercial and widely grown clone in Mbale and Kapchorwa districts of the eastern part of Uganda. It grows better in the highlands than at lower altitudes. K4 was more similar to C3 than to C4. C3 (Nakibizzi) is an introduced clone in the central region, probably from the eastern part, and it was named Nakibizzi (fat pig) due to the large truncated bunches. The values of the correlation and distance coefficients between K4 and C3 were in the same range as those between pairs of known ramets. K4 and C3 were also identical in their qualitative characters, so were regarded as representing a single clone. C4 has not performed well at Kawanda from the time it was introduced into the collection. The suckers which were planted were probably not healthy. The differences between C4 and K4 are therefore thought to be phenotypic rather than genotypic. Mudwale and Nakibizzi are probable synonyms.

Table 6.6: Correlation and distance coefficients between pairs of accessions which shared the same name and other accessions to which they were equally or more similar.

Accessions which shared names (figures in parentheses show differences inquantitative/ qualitative characters)	Correlation with other accessions	Distance from other accessions	Differences in quantitative/ qualitative characters	Conclusion
Musakala clone set				
C2,K2(10/0)	C2, C5 = 0.955	C2, C5 = 0.070	12/0	all members
Corr. 0.925	K2, C5 = 0.925	K2, C5 = 0.091	12/0	of a single clone
Dist. 0.093	C2, K145 = 0.962	C2, K145 = 0.065	11/0	
	K2, K145 = 0.981	K2, K145 = 0.046	11/0	
C4, K4 (11/0)	K4, C3 = 0.922	K4, C3 = 0.099	13/0	C3, C4, K4
Corr. 0.814	K4, C7 = 0.831	K4, C7 = 0.143	12/3	a single clone;
Dist. 0.160				C7 different clone
Nakabululu clone set				
C50, K50 (9/0)	C50, C48 = 0.960	C50, C48 = 0.084	12/2	C49, C50, K50
Corr. 0.949	C50, C49 = 0.964	C50, C49 = 0.081	10/0	a single clone;
Dist. 0.095	K50, C49 = 0.958	K50, C49 = 0.084	10/0	C48 different clone
C51, K51(10/0)	C51, K120 = 0.977	C51, K120 = 0.067	11/0	all members
Corr. 0.929	K51, K121 = 0.944	K51, K121 = 0.104	12/0	of single clone
Dist. 0.119				
Nakitembe clone set				
C19, K19 (10/0)	C19, C20 = 0.867	C19, C20 = 0.131	13/1	C20 and K81
Corr. 0.857	C19, K73 = 0.863	C19, K73 = 0.131	12/0	probably different
Dist. 0.136	C19, K81 = 0.869	C19, K81 = 0.134	12/1	from C19, K19
	K19, C20 = 0.864	K19, C20 = 0.132	13/1	and K73
	K19, K73 = 0.927	K19, K73 = 0.097	13/0	a single clone
	K19, K81 = 0.898	K19, K81 = 0.116	13/1	
C20, K20(12/0)	C20, C21 = 0.900	C20, C21 = 0.115	12/0	C20, K20 C21,
Corr. 0.830	C20, K73 = 0.848	C20, K73 = 0.140	12/1	K21, K81
Dist. 0.154	C20, K81 = 0.912	C20, K81 = 0.109	13/0	members of a
	K20, C21 = 0.904	K20, C21 = 0.117	12/0	single clone
	K20, K73 = 0.865	K20, K73 = 0.140	13/1	and probably
	K20, K81 = 0.912	K20, K81 = 0.108	13/0	different from K73
C22, K22 (11/0)	C22, K77 = 0.966	C22, K77 = 0.088	13/0	all members of
Corr. 0.866	K22, K77 = 0.891	K22, K77 = 0.153		a single clone
Dist. 0.949				

K4 was also more similar to C7 than to C4 (Table 6.6). However, C7 differed from K4 in two of the qualitative characters. C7 had rectangular bunches with lanceolate male buds while K4 had truncate bunches with cordate male buds. C7 was therefore a different clone from K4.

6.4.2.1.2 Nakabululu clone set

In this clone set, C50 and K50 were most similar to C49. C50 was also more similar to C48 than to K50 (Table 6.6). C50 and K50 (Kazirakwe) are clones grown in the south western highlands. C49 (Nakayonga) is grown in the central region while C48 (Nakasabira) is an eastern clone. The names Kazirakwe and Nakayonga, though in different dialects, both mean black coloured or dark coloured. This is because the clones are characterised by intense black pigmentation in the pseudostem up to the petioles. C48 has less intense black pigmentation, along its pseudostem only, but has anthocyanin pigmentation along its upper sheaths, petioles and midribs which is lacking in C49, C50 and K50. C50 and K50 were identical to C49 in their qualitative characters but differed from C48 (Table 6.6). So C50, K50 and C49 are probably the same clone while C48 is a different clone. Kazirakwe and Nakayonga are probable synonyms.

C51 and K51 had their greatest similarities with K120 and K121 respectively (Table 6.6). C51 and K51 (Butobe) originate from the south western parts of Uganda. K120 (Kafunze) originates from the central part, while K121 (Wekhanga) originates from the east. Butobe and Kafunze are old clone names in their respective places of origin. The meaning of the names relates to the narrow and compact bunches. However, the name Wekhanga indicates something new or something which has changed. This means this clone may have been recently introduced in the east. C51 and K51 were identical to K120 and K121 in their qualitative characters (Table 6.6). We therefore assume that C51, K51, K120 and K121 represent a single clone. The names Butobe, Kafunze and Wekhanga therefore become probable synonyms.

6.4.2.1.3 Nakitembe clone set

C19 and K19 (Nakitembe) are accessions believed by farmers to represent the oldest clones of the East African Highland bananas in Uganda and to have given rise to the new clones. All accessions related to C19 and K19 have a characteristically hanging male inflorescence rachis with persistent flowers and a pronouncedly imbricate male bud. Several clones resemble C19 and KI9 so much that farmers name them as variants of Nakitembe. C20 and K20 (Nakitembe-Nakamali or small Nakitembe) and C22 and K22 (Nakitembe-Nakawere or glossy Nakitembe), are part of this complex. Nakawere is a name given to a woman who has just given birth to a child. Nakaweres always look weak and fragile but somehow glossy hence the name Nakitembe-Nakawere. Farmers say that Nakitembe-Nakamali and Nakitembe-Nakawere are mutants derived from C19 and K19.

The name Nakitembe (C19/K19) can mean “being similar to Ekitembe”, the wild banana (Ensete sp.), but the name also originates from the verb “Okutembuka” meaning to hurry. The clone matures very fast, hence the name. In the eastern part of Uganda Nakitembe is called Malira (Lusoga dialect), a name which also relates to the quick maturity of the clone. In the same region there are also Malira-Omutono (Nakitembe-Nakamali or Nakitembe the small type) and Malira-Luvuta (Nakitembe-Nakawere or Nakitembe the glossy one). Omutono and Luvuta also mean “small” and “glossy” respectively.

C19 and K19 are probably ramets. They were identical in their qualitative characters and their pairwise correlation and distance coefficients were in the range of values for ramets. K19 was maintained in the eroded block of the Kabanyolo collection, hence its difference.

C19 was more similar to K81 than K19 while K19 was more similar to K73 than C19 (Table 6.6). The relationship between C19 and K81 has already been discussed indicating that K81 may be a small statured mutant of K19. Simmonds (1966) considered small stature to arise by mutation in other bananas. K73 (Entaragaza) is similar to C19 and K19 but grown in the south western highlands. It has a different name (Entaragaza) because of the different dialect (Runyankore) in that region. The meaning of the name again relates to rapid maturity. K73 is vigorous like C19 and K19. K73 was identical to C19 and K19.

In the section 6.4.1.1 we concluded that C20/K20, the known ramets C21/K21 and K81 were probably all members of the same clone. C20 and K20 differed in 12 of the 13 quantitative characters. K20 was in the second eroded block of the Kabanyolo collection and being naturally a weak clone, it was badly affected by the poor growing conditions, hence its differences from C20. But C20 and K20 represent a single clone.

C22 and K22 (Nakitembe-Nakawere) were more similar to K77 (Luvuta from the eastern part) than to one another. The name Malira-Luvuta was shortened in the Kabanyolo record to Luvuta. K22 was growing in the eroded block of the Kabanyolo collection while K77 was growing in the better maintained part of the same collection when the data were recorded. C22, K22 and K77 probably represent a single clone since the differences between C22 and K22 are likely to have been due to the growing conditions. Nakitembe-Nakawere and Malira-Luvuta become probable synonyms.

6.4.2.2 Accessions which probably represent different clones

Finally, members of five pairs of accessions (C6/K6, C9/K9, C16/K16, C31/K31 and C34/K34) shared the same name but had pairwise distance and correlation coefficients that were outside the ranges of the known ramets (Table 6.7). Some of these accessions were more similar to other accessions in the analyses than to their counterpart accessions (Table 6.7). These accessions belonged to either the Musakala or the Nfuuka clone sets.

6.2.2.2.1 Musakala clone set

In the Musakala clone set, C6 and K6 were both labelled Mayovu but differed in their bunch characteristics. K6 did not match the characteristics of the Mayovu grown by farmers, so K6 had presumably been mislabelled. K6 had an oblique bunch with medium sized fruits with apices of intermediate shape and an oblique male inflorescence rachis. C6 had a pendulous bunch with long fruits with bottle-necked apices and a pendulous male inflorescence rachis.

C6 was more similar to C2, K2, C5, C7, C11, K141, K143, K145 and K146 than to K6, with correlation and distance coefficients within range of known ramets (Table 6.7). C2 (which was shown in section 6.4.2.1.1 page 184 to be the same clone as K2, C5, and K145) differed from C6 by having fruits which were less strongly recurved in the bunch. C11 differed from C6 by having fruits which were strongly recurved to touch the rachis. C6 was identical to C7 (Kisansa), K141 (Mpologoma), K143 (Lwewunzika), all from the central region, and K146 (Rwabakongo) from the western region in all the qualitative characters. Differences between them were in the quantitative characters. They most probably represent a single clone. Mayovu, Kisansa, Mpologoma, Lwewunzika and Rwabakongo become probable synonyms.

Table 6.7: Correlation and distance coefficients between pairs of accessions which shared names but probably represented different clones and those accessions to which they were more similar than to their counterpart accessions.

Accessions sharing names (figure in parentheses shows differences in quantitative/ qualitative characters	Correlation with other accessions	Distance from other accessions	Differences in quantitative / qualitative characters	Conclusion
C6, K6 (12/5)	C6, C2 = 0.890	C6, C2 = 0.112	10/2	C2, K2, C5, K145 represent
Corr. 0.364	C6, K2 = 0.848	C6, K2 = 0.130	10/2	a single clone.
Dist. 0.236	C6, C5 = 0.951	C6, C5 = 0.074	12/2	C6, C7, K141, K143, K146
	C6, C7 = 0.918	C6, C7 = 0.095	13/0	are another clone. K6 mislabelled.
	C6, C11 = 0.892	C6, C11 = 0.108	11/2	C11 is probably different
	C6, K141 = 0.859	C6, K141 = 0.122	12/0	from the rest.
	C6, C143 = 0.911	C6, C143 = 0.099	12/0	K6 probably a single clone
	C6, K145 = 0.893	C6, K145 = 0.108	12/2	with K98.
	C6, K146 = 0.844	C6, K146 = 0.131	13/0	
	K6, K98 = 0.507	K6, K98 = 0.147	13/0	
C9, K9 (11/5)	C9, C10 = 0.759	C9, C10 = 0.142	12/1	C10, C12 are probably different
Corr. 0.400	C9, C12 = 0.875	C9, C12 = 0.104	11/1	clones from C9. C9 and K144
Dist. 0.279	C9, K144 = 0.783	C9, K144 = 0.139	13/2	are both different clones from
	K9, C16 = 0.740	K9, C16 = 0.201	13/0	one another and from C9. K9 is probably the same clone as C16.
C16/K16 (12/5)	C16, KM15 = 0.963	C16, KM15 = 0.066	13/0	C16, KM15 are a single clone.
Corr. 0.313	K16, K85 = 0.800	K16, K85 = 0.091	11/2	K16 (mislabelled) is a different
Dist. 0.239				clone from C16 and from K85.
C31, K31(12/0)	C31, C37 = 0.783	C31, C37 = 0.075	12/0	Difficult to tell whether K31 is
Corr. 0.208	C31, C38 = 0.799	C31, C38 = 0.077	13/1	different from C31 or they are
Dist. 0.166	C31, K38 = 0.868	C31, K38 = 0.058	9/1	the same (correlation and distance
	C31, K94 = 0.930	C31, K94 = 0.044	11/0	values too low). C31, C37, K94,
	C31, K99 = 0.798	C31, K99 = 0.077	13/0	and K99 represent a single clone. C38 and K38 are different from the rest.
C34, K34 (11/0)	K34, K152 = 0.966	K34, K152 = 0.058	13/0	Single clone
Corr. 0.666				
Dist. 0.198				

The reason that this clone C6, has several names in the central region alone is probably because it originated repeatedly and independently. It is believed however, to have been introduced in the central region from the east and farmers obtained it from different sources with no name. At one time suckers were being supplied by agricultural offices at the district centres, as a clone most favourable for the market. K6 was more similar to K98 than to C6 but the correlation coefficient with K98 was not within the range of values of ramets (Table 6.7). K6, however, was identical in the qualitative characters scored with K98. K6 and K98 could therefore represent a single clone. The fact that K6 was growing in an eroded block could have been responsible for low coefficient values, otherwise this is another example where the two coefficients interpreted data very differently.

Two other accessions formerly and subjectively classified as members of Musakala clone set, C16 and K16, were both labelled Atwalira but differed so much that K16 was considered to have been mislabelled. C16 was similar to C15/K15 except that it lacked red petioles and midribs. In C16, the red colouring is only found in marginal areas of the petiole and upper sheaths. Farmers considered C16 to have evolved from C15/K15. C16 was identical to KM15 (Siira White), a mutant of K15 which evolved in the Kabanyolo collection, in the qualitative characters but differed in all the thirteen quantitative characters (Table 6.7). C16 and KM15 probably represented a single clone. Atwalira, Siira White are probable synonyms. Siira White is a name which was given to a mutant of Siira in the Kabanyolo germplasm collection.

K16, on the other hand was more similar to K85 (Entazinduka) a clone from the south western highlands (Table 6.7). K16, however, had oblique rectangular bunches whereas Entazinduka had pendulous cylindrical bunches, so the two are considered to be different clones. K16 was also similar to a number of accessions in the Nfuuka clone set, notably K94 (Enkobe from west Uganda) and K99 with distance coefficients which were in the range for ramets but correlation coefficients which were slightly below the range for ramets. K16 was identical to K94 and K99 in the qualitative characters and was hence considered to belong to the same clone. K99 has already been shown to belong to the same widespread clone as the ramets C37/K37, and K95 and K98 (and now probably K6). The values of the distance coefficients between K16, K94, and C37/K37, K95, K98 support the conclusion that they are all one clone. Enkobe (K94) therefore becomes another synonym of Enyeru (C37/K37).

C9 and K9 were both labelled Nalugolima, but differed in features of bunch, fingers and latex. The name Nalugolima is used by farmers for two different clones, with characters of C9 and K9 respectively. The two accessions entered the collections independently, from different sources and the differences between them are not due to mistakes in curation. Although C9 and K9 were both collected from the central region, K9 is considered to have been introduced to the central region.

C9 was similar to two other accessions in the Musakala group: C10 (Muturit) and C12 (Mugisu-agenda), both from the eastern region (Table 6.7). However, C10 and C12 have a shorter peduncle than C9 (30cm as opposed to 40) and their bunches are held up close to base of the peduncle. Peduncle length/girth ratio, thus differed between the three clones, C9 having a larger ratio than the two other accessions. C9 may represent a different clone from C10 and C12.

C9 was also found to be similar to K144 (Mujuba) from the western highlands but differed in two qualitative characters and hence was considered a different clone from K144 (Table 6.7).

K9 was more similar to the Siira accessoins (C15, K15, KM15, C16, K90) than to the Musakala group. K9 was identical to KM15, C16 and K90 (Nassaba) in the qualitative characters, and it had its greatest similarity (and least dissimilarity) with C16 (Table 6.7), but the correlation and distance coefficients were not within the ranges of values of ramets. K9 was maintained in the eroded block of Kabanyolo collection and this could have caused differences between K9 and the three Siira accessions. K9 is probably the same clone as KM15, C16 and K90. Nalugolima is therefore probably an additional synonym of Siira White and Atwalira.

6.2.2.2.2 Nfuuka clone set

C31 and K31 (Nfuuka) are accessions on which one of the names of the subjective classification is based (Nfuuka means “I am changing” or “I keep changing”). These, together with C34/K34 (Ndyabalangira), are some of the original accessions reported by farmers to change readily. Accessions in each pair differed greatly from one another in their quantitative characteristics but were identical in their qualitative characters. K31 and K34 were located in the first two blocks in the Kabanyolo collection which were eroded and plants in these two blocks were not growing well. The blocks were not mulched and the ground was not fertile. It is likely that the accessions were greatly affected by the growing conditions.

It is very difficult to decide whether C31 and K31 represented a single clone. This is because their pairwise correlation between C31 and K31 was too low (distance too large) to consider the two to be the same clone (Table 6.5). K31 had its highest similarity (least dissimilarity) with C35 (Tuula-twogere) but the values were well below those of ramets. However, C31 and K31 were identical in their qualitative characters which were scored and this suggests that they were probably a single clone but K31 was greatly affected by the growing condtions.

C31 (Nfuuka) was so similar to K94 (already identified as the same clone as K16) and K99 (identified the same clone as C37/K37, K16, K94, K95, K98) that these 10 accessions are considered to belong to a single clone. Nfuuka, become an additional synonym of Enyeru. C31 was also found to be more similar to C38/K38 (Nakhaki) than to K31. C38/K38 differed from C31 in the colour of fruit pulp before and after maturity. So C31 was different from C38/K38.

C34 and K34 differed greatly in half of the 13 quantitative characters. C34 was nevertheless more similar to K34 than to any other clone in the analysis. K34 was more similar to K152 (Enzirabushera) than to C34 (Table 6.7). K152 is from the south western part of the country. The three accessions, C34, K34 and K152 are the only accessions among the Highland banana clones with chocolate brown pigments in their pseudostems. K34 and K152 were growing in the eroded block of the Kabanyolo collection. The three accessions (C34, K34, K152), however, were identical in their qualitative characters and were considered to represent a single clone. At the end of this phase, it is important to note that phenotypic duplicates should not be discarded solely on the basis of a shared local name. Critical studies are necessary to confirm whether such accessions are similar or different (Pickersgill, 1994; Karamura et al., 1996).

6.4.3 Similar accessions which do not share the same name

Further reference to both distance and correlation matrices indicated that a number of accessions which had different names were likely to belong to the same clone. These accessions will be discussed in relation to the clone sets to which they were assigned.

6.4.3.1 Beer clone set

Table 6.8 shows the beer accessions which were sufficiently similar to be possibly members of the same clone but which had different names. KB128 (Enywamaizi) was found to be similar to CB53 and KB53 (Namadhi from the eastern part of the country). KB128 is a clone widely grown in the south western highlands and one of the oldest beer clones in Uganda. It is distributed in all banana growing areas. In the

central region it is called Kabula. Kabula seems to be disappearing in the central region where farmers are growing instead the introduced ABB beer types.

Kabula is the name of the county in the western part of Uganda where the clone used to be widely grown. But the name also relates to the strong drink it produces. The strong drink may be related to the high levels of condensed tannins found in the fruit pulp (Aked, 1995). The names Namadhi and Enywamaizi relate to the word water meaning the abundant sap found in the fruits. The three accessions (CB53, KB53, KB128) were identical in their qualitative characters but differed in their quantitative characters (Table 6.8). They probably represent a single clone and farmers' information supports this. Namadhi and Enywamaizi are probable synonyms. Two other names have the same meaning as Namadhi and Enywamaizi. These are Nametsi and Nalusi, but the two accessions bearing these names were different from Namadhi and Enywamaizi. Namadhi, the name to be used for this clone, is a clone representative of the Beer accessions. This is because it is considered to be the oldest Beer clone known by the farmers.

Table 6.8: Pairs of accessions in the Beer clone set with correlation and distance coefficients within the range of ramets.

Accessions	Correlation coefficient	Distance coefficient	Quantitative/ qualitative character differences	Conclusion
CB53/KB128	0.907	0.117	10/0	All one clone
KB53/KB128	0.921	0.107	10/0	
CB57/KB56	0.949	0.080	11/0	All one clone
KB57/KB56	0.956	0.075	10/0	
CB57/KB132	0.902	0.114	11/0	
KB57/KB132	0.912	0.109	13/0	
KB132/KB56	0.868	0.133	13/0	
KB133/KB57	0.806	0.149	13/0	
CB57/KB87	0.806	0.056	11/2	Different clones
KB57/KB87	0.769	0.141	12/2	
KB123/KB124	0.986	0.048	11/0	All one clone
KB123/KB125	0.976	0.064	13/0	
KB123/KB126	0.976	0.061	11/0	
KB124/KB125	0.969	0.073	12/0	
KB124/KB126	0.979	0.058	13/0	
KB125/KB126	0.904	0.077	10/0	
CB54/KB137	0.795	0.140	13/3	Different clones
CB54/KB159	0.854	0.125	13/3	
KB137/KB159	0.883	0.097	13/0	Single clone
CB55/KB135	0.896	0.110	12/3	Different clones
CB55/KB136	0.884	0.116	12/3	
KB135/KB136	0.971	0.054	13/0	Single clone
KB158/KB159	0.903	0.089	12/0	All one clone
KB158/KB131	0.915	0.083	11/0	
KB158/KB134	0.873	0.104	13/0	
KB158/KB138	0.915	0.084	13/0	
KB158/KB139	0.850	0.112	13/0	
KB158/KB140	0.919	0.082	13/0	

Table 6.8 (Cont'd.)

Accessions	Correlation coefficient	Distance coefficient	Quantitative/ qualitative character differences	Conclusion
KB158/KB49	0.806	0.128	13/0	
KB131/KB134	0.884	0.098	13/0	
KB131/KB138	0.938	0.070	12/0	
KB131/KB139	0.913	0.085	12/0	
KB131/KB140	0.829	0.123	13/0	
KB131/KB49	0.939	0.070	13/0	
KB134/KB138	0.971	0.050	12/0	All one clone
KB134/KB139	0.856	0.111	11/0	
KB134/KB49	0.887	0.089	13/0	
KB134/KB140	0.859	0.109	13/0	
KB138/KB139	0.869	0.104	12/0	
KB138/KB49	0.892	0.094	11/0	
KB138/KB140	0.919	0.081	13/0	
KB139/KB49	0.951	0.065	13/0	
KB139/KB140	0.904	0.089	13/0	
KB49/KB140	0.879	0.100	13/0	
KB130/KB158	0.791	0.146	13/1	Different clones
KB130/KB131	0.867	0.117	12/1	
KB130/KB49	0.831	0.122	13/1	
KB133/KB131	0.827	0.132	13/2	Different clones
KB133/KB158	0.772	0.142	13/2	
KB133/KB134	0.786	0.139	12/2	
KB133/KB138	0.791	0.136	13/2	
KB133/KB139	0.829	0.123	12/2	
KB133/KB49	0.843	0.120	12/2	
KB133/KB140	0.794	0.134	13/2	
CM4/KB84	0.764	0.145	13/0	Single clone

Three accessions, KB56 (Enyarukira from the west), KB87 (Namunyere from the east) and KB132 (Shombo-obureku from the south west) were similar to CB57 and KB57 (Nalukira) in having pendulous cylindrical bunches with a very long inflorescence rachis. The name Nalukira or Enyarukira relates to the long tail; indicating the long male inflorescence rachis. KB56 and KB132 were identical to each other and to CB57 and KB57 (Table 6.8) in all the qualitative characters and they probably represent a single clone. Nalukira, Enyarukira and Shombo-obureku are probable synonyms. KB87 however, differed from CB57 and KB57 in having more compact bunches of smaller fruits arranged almost at right angles to the rachis, an inflorescence rachis which was oblique, not pendulous like the four other accessions, and a style which was straight, not hooked as in the others. KB87 was therefore a different clone from the others.

KB133 was identical to KB56, KB57, CB57 and KB132 in all the qualitative characters but differences existed in all the quantitative characters. The values of the correlation coefficient support this identity because they were within range of

ramets but not those of distance coefficient. This could be the effects of distance coefficient on size differences. KB133 represented a single clone with KB56, CB57, KB57 and KB132.

Four other accessions, KB123 (Enkara), KB124 (Enshyenyuka), KB125 (Entanga) and KB126 (Imbululu), were identical to each other in their qualitative characters (Table 6.8). The first three accessions originate from the south western region while KB126 originates from the eastern region. The 4 were considered to represent a single clone (Table 6.8). Enkara, Enshyenyuka, Entanga and Imbululu are therefore probable synonyms.

CB54 (Mende), a beer clone only known in Luweero district, central region was found to be similar to KB137 (Kaitaluganda) and KB159 (Naminwe) both from the eastern part of Uganda. However, KB137 and KB159 differed from CB54 by having rectangular fruits with apices of intermediate shape while CB54 had rounded blunt fruits. KB137 and KB159 were however, identical in all their qualitative characters and probably represent a single clone (Table 6.8). It is important to note that the name Naminwe relates to few fingers in the bunch. But KB159 did not show few fingers. Kaitaluganda therefore can not possibly be synonymous with Naminwe although accessions KB137 and KB159 represent a single clone. KB159 was probably mislabelled.

CB55 (Bagandeseza), another clone known only in Luweero district, central region was found to be similar to KB135 and KB136. KB135 (Enjumba) and KB136 (Engambani), are two common beer clones from the south western part of Uganda. CB55 differed from KB135 and KB136 in bunch characters and in most of the quantitative characters. KB135 and KB136, on the other hand, were both identical in their qualitative characters but had minor differences in all their quantitative characters (Table 6.8). They may represent a single clone. This makes Enjumba and Engambani probable synonyms. It is alleged by farmers that Entukura beer which is not represented in the collection is the same clone as KB135 and KB136.

Table 6.8 further suggests that KB158 (Mwanga from the west) was similar to KB159 (which has been found to be the same clone as Kaitaluganda KB137), KB130 (Nalusi from the east), KB131 (Liyase from the east), KB133 (Kibagampera from the east), KB134 (Engumba from the central region), KB138 (Nalwesanya from the central region), KB139 (Endembezi from the south west), KB140 (Mutant-mixed from the south west) and KB49 (Nakayonga from the central region). KB158 was identical in all the qualitative characters that were scored to 7 of these 9 accessions but differed from KB130 and KB133.

Results in Table 6.8 further indicate that the 7 accessions which were identical to KB158 were also identical to each other and like KB158, differed from KB130 and KB133. These 7 accessions are considered to belong to the same clone. The names Mwanga, Kaitaluganda, Liyase, Engumba, Nalwesanya, Endembezi and Mutant-mixed therefore become probable synonyms.

KB130 (Nalusi from the east) was a small statured clone while the 7 accessions and KB133 were tall clones. KB133 had cylindrical bunches different from those of the 7 accessions and KB130. Hence KB130 and KB133 were different from the 7 accessions and also different from each other (Table 6.8).

The name Nalusi relates to flowing water like Namadhi (CB53, KB53 and KB128) and these two clones were considered by farmers to be the same. However, the fruits

of Nalusi were not rounded or blunt like those of Namadhi. The values of correlation and distance coefficients also do not suggest that KB130 is the same as CB/KB53 and KB128.

Two beer accessions, CM4 (Mudwale beer from the east), claimed by the farmers to be a mutant of C4, and KB84 (Oruhuuna from the south west) had been placed in the Nakitembe clone set by cluster analysis. However, discriminant analysis placed them back to their subjective clone set. They were identical in all qualitative characters, but differed in all 13 quantitative characters (Table 6.8). The two were considered to represent the same clone.

6.4.3.2 Musakala clone set

K1 and C1 (Muvubo from the central region) were identical to K144 (Mujuba from the south west) (Table 6.9) in all their qualitative characters. They however differed greatly in the quantitative characters. K144 was growing in the eroded part of the Kabanyolo collection and thus probably differed from K1 and C1. The three were considered to represent a single clone.

C4/K4 and C3 were also identical to C13/K13 in all the qualitative characters. The estimation of resemblance by correlation coefficient between C4/K4/C3 and C13/K13 was within range of the value of ramets but not distance coefficient. C13 and K13 are western Uganda clones which had been introduced there from the East. The two clones are most probably the same as C4/K4 and C3. The values of the distance were below the range of values for ramets. This is probably another example where the distance coefficient was interpreting data differently from that of the correlation coefficient, since all these accessions were growing in good conditions and none was in the eroded blocks of the Kabanyolo collection.

Table 6.9: Pairs of accessions in the Musakala clone set with correlation and distance coefficients within the range of ramets

Accessions	Correlation coefficient	Distance coefficient	Quantitative/ qualitative character differences	Conclusion
C1/K144	0.773	0.147	13/0	All one clone
K1/K144	0.797	0.148	13/0	
C3/C13	0.786	0.159	13/0	Single clone
C4/C13	0.759	0.196	13/0	
K142/C2	0.867	0.122	12/1	K142 is different
K142/K2	0.819	0.143	13/1	clone from C2/K2
K142/C6	0.848	0.130	13/0	Single clone
K142/C11	0.879	0.113	13/1	Different clones
C9/C6	0.775	0.142	12/0	Single clone
C12/C10	0.866	0.113	13/0	Single clone
C12/C13	0.812	0.135	13/2	Different clones
C12/K13	0.813	0.134	12/2	
C12/K103	0.772	0.141	13/4	

K142 (Rwamigongo from the west) was similar to C2 (Musakala, a name which already has two synonyms), C6 (Mayovu, another name with four synonyms) and C11 (Lumenyamagali from the central region) (Table 6.9). K142 was identical to C6, and to all other accessions considered to belong to this clone in all the qualitative characters, but differed from C2 in the orientation of the fruits relative to the rachis. Rwamigongo therefore joins the list of synonyms of Mayovu. C11 differed from K142 and other members of the Mayovu clone by having strongly recurved fruits almost touching the rachis. Lumenyamagali was thus considered distinct from Mayovu.

C9 (Nalugolima) was similar to C6 (Table 6.9). They were identical in all the qualitative characters although they differed in their quantitative characters. C9 and C6 represent a single clone.

C12 was similar to C10 (Muturit), C13 and K13 (Wansimirahi from the west) and K103 (Enyamanshyari from the west) (Table 6.9). However, C13 and K13 differed from C12 by having truncated pendulous bunches and a characteristically cordate male bud. K103 lacked the slender fruits of C12. Instead it had triangular fruits. K103 was therefore different from C12. C12 was identical to C10 in all the

qualitative characters and hence considered to be the same clone as C10. Muturit and Mugisu-agenda are probable synonyms.

6.4.3.3 Nakabululu clone set

In this clone set accessions K28 (Ekitetengwa from the west) was similar to C27 and CM27. However, K28 differed from the other two by having persistent floral parts along its inflorescence rachis. These are accessions which show resemblance to Nfuuka and Nakitembe clone sets. They had been subjectively classified as Nakitembe accessions and discriminant analysis placed here.

K118 (Bifusi from the east) and K119 (Embururu from the east) were similar to C47 and K47 (Nakabululu from central region) (Table 6.10). However, K118 differed from the other three by having inflated fruits. C47, K47 and K119 were identical in all their qualitative characters and differed only in their quantitative characters. C47, K47 and K119 represented a single clone but K118 was different (Table 6.10). Nakabululu and Embururu are probable synonyms.

K122 (Mukite from the east) was found to be identical to both C48 and K48 (Nakasabira from the east) (Table 6.10). Although both originate from the east,

they grow in different localities. Mukite and Nakasabira are probable synonyms.

6.4.3.4 Nakitembe clone set

A number of accessions were found to be similar in the Nakitembe clone set (Table 6.11). C24 (Nakitembe-red), a clone from the central region, resembled K78 (Nalwela), a clone from the eastern region. Both accessions had anthocyanin pigmentation along the upper sheaths, petioles and midribs. However, C24 was a robust plant whereas K78 was small. The two accessions differed considerably in the quantitative characters. K78 was in an eroded block at Kabanyolo and was affected by the growing conditions. C24 and K78 were identical in all qualitative characters which were scored and they probably represented a single clone.

Table 6.10: Pairs of accessions in the Nakabululu group with correlation and distance coefficients within the range of ramets

Accessions	Correlation coefficient	Distance coefficient	Quantitative/ qualitative character differences	Conclusion
C27/K28	0.903	0.095	13/2	Different clones
CM27/K28	0.905	0.094	12/2	
K118/C47	0.906	0.115	13/1	K118 is
K118/K47	0.777	0.140	13/1	different from,
K118/K119	0.875	0.136	13/1	C47/K47, K119
K119/C47	0.829	0.141	12/0	Single clone
K119/K47	0.946	0.088	13/0	
K121/C52	0.901	0.146	13/2	Different clones
K121/K52	0.904	0.137	13/2	
K122/C48	0.869	0.145	13/0	All one clone
K122/K48	0.854	0.147	13/0	

Table 6.11: Pairs of accessions in the Nakitembe group with correlation and distance coefficients within range of ramets

Accessions	Correlation coefficient	Distance coefficient	Quantitative/ qualitative character differences	Conclusion
C24/K78	0.951	0.085	13/0	Single clone
C25/K149	0.977	0.083	12/0	Single clone
K148/K150	0.884	0.139	13/1	Different clones
K72/K74	0.950	0.088	13/0	All one clone
K72/K75	0.928	0.104	12/0	
K72/K76	0.906	0.117	13/0	
K74/K75	0.962	0.072	11/0	
K74/K76	0.940	0.091	13/0	
K75/K76	0.968	0.067	13/0	
K79/K80	0.909	0.126	13/0	Single clone
K82/C18	0.916	0.143	13/0	All one clone
K82/K18	0.927	0.147	13/0	
K82/K83	0.990	0.056	12/0	
K82/K84	0.963	0.107	13/0	
K83/C18	0.923	0.145	13/0	
K83/K18	0.933	0.144	13/0	
K83/K84	0.953	0.122	11/0	
K84/C18	0.881	0.146	13/0	

C25 (Nakibule) from the central region was similar to K149 (Waikova) from the eastern region. They had intense bronze to black pigmentation in the pseudostems, along the petioles and midribs. They are both small statured. C25 and K149 were identical in all the qualitative characters (Table 6.11). They represented a single clone.

Accessions K148 (Enyarutere from the west) and K150 (Bikowekowe from the east) were very similar to each other. However, K150 had fruits which were rounded with blunt tips and arranged almost horizontally to the rachis. Fruits in K148 were rectangular in shape with intermediate shaped apices. The two therefore were different clones (Table 6.11).

Several accessions in the Nakitembe clone set possessed pronounced curved angular fruits and a pronounced fresh style. These accessions were K72 (Nandigobe from the central region), K74 (Enjagata from the western region), K75 (Toro from the west), K76 (Ntinti from the east), K79 (Nabuyobyo from the east) and K80 (Nasaala also from the east). The first four were identical in all their qualitative characters. They represented a single clone (Table 6.11), although originating from different regions. Nandigobe, Enjagata, Toro and Ntinti are probably synonyms. K79 and K80 were from the eastern highlands and they looked like small statured types of K72, K74, K75 and K76. Their pairwise correlation coefficients with the members of the Enjagata clone were within range of ramets but their distance coefficients were below the values of ramets. K79 and K80 were identical in the qualitative characters and hence considered to represent a single clone. Nasaala and Nabuyobyo are probably synonyms.

Another group of similar accessions within the Nakitembe consisted of C18 and K18 (Namaliga from central region), K82 (Enshakara from south west), K83 (Rwakashita from the south west) and K84 (Ebihuuna from the south west). The four were identical in the qualitative characters (Table 6.11), and were considered to represent a single clone.

6.4.3.6 Nfuuka clone set

Several accessions in the Nfuuka group had different names but were phenetically similar. K17 (Kabucuragye) was very similar to K85 and K86. K85 (Entazinduka from the west) and K86 (Enjogabakazi from the west) were both identical in the qualitative characteristics and were considered to represent a single clone (Table 6.12). K17 (Kabucuragye from the west) was probably a different clone.

K30 (Nakinyika) was identical to KM30 (Enyakinika, a supposed mutant of K30) and K96 (Kufuba from the east). Their bunches tend to curve after the first hand

and their names are related to this. They represent a single clone (Table 6.12).

C37 (Enyeru, a commercial south western clone) was chosen to represent the accessions which have already been considered to represent the same clone (section 6.4.2.2.1). C37 was identical in qualitative characters to CM32 (Namwezi Black from the central region), C41 (Enjeriandet) an eastern clone, K157 (Enyaruyonga from the west) and K97 (Rwasha from the west) and K112 (Khabusi from the East) (Table 6.12). They probably represented a single clone growing in different regions. Enyeru, Namwezi Black, Enjeriandet, Rwasha and Enyaruyonga are probable synonyms.

K93 (Nabusa), K88 (Kasitaza), K89 (Kisabo) and K92 (Mutta-Ngendo) were identical in their qualitative characters (Table 6.12). All four accessions are from the

Table 6.12: Pairs of accessions in the Nfuuka clone set with correlation and distance coefficients within range of ramets

Accessions	Correlation coefficient	Distance coefficient	Quantitative/ qualitative character differences	Conclusion
K17/K85	0.780	0.092	11/2	Different clones
K17/K86	0.813	0.085	12/2	
K85/K86	0.861	0.075	11/0	Single clone
K30/KM30	0.768	0.099	13/0	All members of a
K30/K96	0.841	0.075	13/0	single clone
C37/CM32	0.791	0.090	13/0	All members
C37/K157	0.788	0.081	13/0	of a single clone
K37/K97	0.762	0.131	13/0	
K37/K112	0.762	0.089	10/0	
K88/K89	0.823	0.096	12/0	All members
K88/K92	0.919	0.065	12/0	of a single clone
K89/K92	0.837	0.091	12/0	
K92/K93	0.801	0.104	8/0	
K90/KM15	0.835	0.146	13/0	Single clone
K102/K99	0.774	0.089	10/1	Different clones
K33/C32	0.906	0.117	10/2	Different clones
C40/K153	0.963	0.076	9/0	All one clone
K153/K154	0.950	0.087	11/0	
K107/K108	0.795	0.113	13/0	Single clone
K107/K109	0.799	0.123	13/2	Different clones
K108/K109	0.822	0.122	13/2	
K110/K113	0.829	0.103	13/2	Different clones
K111/K114	0.943	0.070	11/0	Single clone
K115/K116	0.867	0.143	12/0	Single clone
K115/K117	0.885	0.131	11/0	Single clone
K116/K117	0.905	0.123	12/0	Single clone
C46/K104	0.918	0.067	10/0	Single clone
K105/C46	0.922	0.070	13/0	Single clone
K105/K104	0.924	0.064	10/0	Single clone
C46/K156	0.866	0.088	12/1	Different clones
K156/K106	0.759	0.123	12/1	

central region. Nabusa is an old and widely distributed clone in the central region. Kasitaza, Kisaabo and Mutta-Ngendo occupy localities where Nabusa was not originally grown. Hence they were given different names but are considered by farmers to be the same as Nabusa. Nabusa, Kasitaza, Kisaabo and Mutta-Ngendo are probable synonyms.

It has been widely known by farmers and traders that Nabusa (K93), a clone which now represents the accessions which have already been considered to represent the same clone (K88, K89 and K92), is the same clone as Enyeru, a commercial clone commonly grown in the western highlands. Enyeru seems to thrive well in that part of the country. In the central region Nabusa does not seem to thrive well. Its bunches tend to be smaller in size. It however, matures very fast and spreads fast and it is tasty a reason which could be attributed to the climatic conditions. This study however, indicate that they may be different. Table 6.13 shows their correlation and distance coefficients which do seem to support that Enyeru and Nabusa are the same. The two accessions were identical in the qualitative characters that were assessed, but differed greatly in their quantitative characters. Nabusa is known to be very sensitive to different growing conditions as indicated by farmers in the central region and it was growing in the first eroded block of the Kabanyolo collection. There is a possibility that these growing conditions could be responsible for the big difference between Enyeru (C37/K37) and Nabusa (K93). The probability of Enyeru and Nabusa to represent a single clone is there. It would however be interesting to look at their differences based on molecular studies.

Table 6.13: Correlation and distance coefficient between C37/K37 and K93.

Accession (Figs. in parenthesis show differences in quantitative and qualitative characters)	Correlation coefficient	Distance coefficient
K93/C37(13/0)	0.398	0.158
K93/C37(13/0)	0.468	0.160

K90 (Nassaba) was identical to KM15 (Siira White)(Table 6.12) which was earlier found to be the same clone as C16 (Atwalira). Nassaba is an additional synonym of Siira White and Atwalira.

K102 (Kabende from the eastern part of Uganda) was similar to K99 in most of the qualitative characters (Table 6.12). However, K102 had variegated fruits, but K99 did not.

K33 (Kulwoni from the east) was found to be similar to C32 and K32 (Namwezi from the central region). These are accessions which lack anthocyanin pigments in the vegetative parts. K33 is semi-dwarf and has a greyed yellow undersheath. The two clones are therefore different (Table 6.12).

Other accessions which shared identical qualitative characters in the Nfuuka group, were C40 (Nakawere from the central region), K153 (Kasenene from the western region) and K154 (Karinga from the west). These also constitute a single clone (Table 6.12).

K107 (Lusumba from the central region) and K108 (Namamuka from the eastern region) were very similar to K109 (Nambi from the central region). However, K107 and K108 have characteristically gourd shaped but slightly inflated fruits whereas K109 has small rounded but inflated fruits. K107 and K108 were identical in all the qualitative characters and were considered to represent a single clone (Table 6.12), but K109 represents a different clone.

K110 (Enyabakazi) and K113 (Nyabungere) were both from the western region and were both similar. K110 is a clone with very compact bunches and with fruits having dark round style scars on their tips or sometimes a persistent dry style. K113 did not have very compact bunches. Its fruits retain fresh styles on their apices. Hence K110 and K113 are different clones (Table 6.12).

K111 (Enyakagongo from the west) was identical in all its qualitative characters with K114 (Namakhumbu from the east). The two accessions were different in the quantitative characters. It is probable that the two represent a single clone making Namakhumbu and Enyakagongo synonyms.

Accessions K115 (Bukumo from the south west), K116 (Enkonera from the western region) and K117 (Kisugunu from eastern region) represented a single clone. They were identical in all their qualitative characters but differed in the quantitative characters. Bukumo, Enkonera and Kisugunu are therefore synonyms.

C46, K104, K105, K156 were accessions with anthocyanin pigments along the uppersheaths, petioles and midribs. K104 (Nambogo or Nambokho) is a very old clone from the east. It was identical to C46 (Namande) and K105 (Nakijumbi). C46 and K105 may have been introduced to certain localities in the central region with no name and farmers named them Namande or Nakijumbi according to the farmers who introduced them. K104, C46 and K106 represented a single clone but originating from different regions (Table 6.12). Nambogo, Namande and Nakijumbi are probable synonyms.

C46 (Namande) and K156 (Kibalawo) from the central region were claimed by some farmers to be different but the differences could only be seen in the quantitative characters. However, K156 had probably a deeper red colour than C46 and it was semidwarf in stature hence both accessions were different clones Kibalawo is an old name which relates to the red traditional cloth worn by women in the central region. It indicates a deep red colouring. The intensity of anthocyanin pigmentation was found to be influenced by environmental conditions. Deep red is associated with wet seasons whereas light red occurs during dry conditions. The time of scoring this character matters.

6.4.4 Probable synonyms and distinct clones

Table 6.14 summarises the synonyms determined after the study of accessions in the two national collections and, the number of distinct clones present in each clone set are summarised in Table 6.15.

Of the accessions which shared the same name, twenty out of twenty two pairs were found to be probable ramets of the same clone. One pair (C16/K16) was considered to contain a mislabelled accession. One pair (C9/K9) represented a name which was applied to two different clones by farmers. At the end of this phase, results indicated that a single clone can have two to ten names. The number of names relate to how widespread the clone is. The wide spread of a clone also depends on the adaptability of a particular clone in the different environment.

Table 6.14: Probable synonyms of the 192 accessions studied from the national collections

Probable synonyms	Region	Name chosen for this clone
Beer clone set		
Namadhi (CB53, KB53)	east	Namadhi
Enywamaizi (KB128)	south west	
Enyarukira (KB56)	south west	Nalukira
Nalukira (CB57, KB57)	central	
Shombo-obureku (KB132)	south west	
Kibagampeera (KB133)	east	
Enkara (KB123)	south west	Entanga
Enshyenyuka(KB124)	south west	
Entanga (KB125)	south west	
Imbululu (KB126)	east	
Enjumba (KB135)	west	Engambani
Engambani (KB136)	south west	
Liyase (KB131)	east	Engumba
Engumba (KB134)	central	
Kaitaluganda(KB137)	east	
Nalwesanya (KB138)	central	
Endembezi (KB139)	south west	
Mutant mixed (KB140)	south west	
Mwanga (KB158)	south west	
KB49	central	
Mudwale Beer (CM4)	east	Oruhuuna Beer
Oruhuuna Beer (KB84)	south west	
Musakala clone set		
Muvubo (C1, K1)	central	Muvubo
Mujuba(K144)	south west	
Musakala (C2, K2)	central	Musakala
Luwata (C5)	central	
Bandageya (K145)	west	
Mayovu (C6)	central	Mayovu
Kisansa (C7)	central	
Nalugolima (C9)	central	
Mpologoma (K141)	central	
Rwamigongo (K142)	west	
Lwewunzika (K143)	central	
Rwabakongo (K146)	west	
Mudwale (C4, K4)	east	Mudwale
Nakibizzi (C3)	central	
Wansimirahi (C13, K13)	south west	
Muturit (C10)	east	Muturit
Mugisuagenda (C12)	east	
Nakabululu clone set		
Nakabululu (C47, K47)	central	Nakabululu
Embururu (K119)	east	
Kazirakwe (K50, C50)	south west	Kazirakwe
Nakayonga (C49)	central	

Table 6.14 (Cont'd.)

Probable synonyms	Region	Name chosen for this clone
Nakabululu clone set (cont'd.)		
Butobe (K51, C51)	south west	Butobe
Kafunze (K120)	central	
Wekhanga (K121)	east	
Mukite (K122)	east	Mukite
Nakasabira (C48, K48)	east	
Nakitembe clone set		
Nakitembe (C19, K19)	central	Nakitembe
Entaragaza (K73)	south west	
Nakitembe-Nakamali C20, K20)	central	Nakitembe- Nakamali
Namulondo (C21, K21)	central	
Nakangu (K81)	east	
Nakitembe-Nakawere (C22, K22)	central	Nakitembe-Nakawere
Luvuta (K77)	east	
Nakitembe red (C24)	central	Nalwela
Nalwela (K78)	east	
Nakibule (C25)	central	Waikova
Waikova (K149)	east	
Nandigobe(K72)	central	Enjagata
Enjagata (K74)	south west	
Toro (K75)	west	
Ntinti (K76)	east	
Nasaala (K80)	east	Nasaala
Nabuyobyo (K79)	east	
Namaliga (C18, K18)	central	Oruhuuna cooking
Enshakara (K82)	south west	
Rwakashita (K83)	south west	
Oruhuuna cooking (K84)	south west	
Nfuuka clone set		
Entukura (C44, K44)	south west	Entukura
Enzirabahima (C45, K45)	south west	
Ndyabalangira (C34, K34)	central	Nzirabushera
Nzirabushera (K152)	south west	
Entazinduka (K85)	south west	Entazinduka
Enjogabakazi (K86)	south west	
Nalugoloma (K9)	central	Nassaba
Siira white (KM15)	central	
Atwalira (C16)	central	
Nassaba (K90)	east	
Nakinyika (K30)	central	Nakinyika
Enyakinika (KM30)	south west	
Kufuba (K96)	east	
Atwalira (K16)	central	Enyeru
Enkobe (K94)	west	
Enjeriandet (C41)	east	
Enyaruyonga (K157)	west	
Enyeru (C37, K37)	west	

Table 6.14 (Cont'd.)

Probable synonyms	Region	Name chosen for this clone
Nfuuka clone set (cont'd.)		
Kasitaza (K88)	central	
Kisaabo (K89)	central	
Khabusi (K112)	east	
Likhago (K95)	east	
Mayovu (K6)	east	
Muttangendo (K92)	central	
Nabusa (K93)	central	
Namwezi black (CM32)	central	
Nfuuka(C31)	central	
Rwasha (K97)	west	
Serunjogi black (K99)	central	
Lusumba (K107)	central	Lusumba
Namamuka (K108)	east	
Nakawere (C40)	central	Kasenene
Kasenene (K153)	south west	
Karinga (K154)	south west	
Enyakagongo (K111)	west	Namakhumbu
Namakhumbu (K114)	east	
Bukumo (K115)	west	Bukumo
Enkonera (K116)	east	
Kisugunu (K117)	east	
Namande (C46)	central	Nambokho
Nambogo (K104)	east	
Nakijumbi (K105)	central/west	

Table 6.15: Number of distinct clones found in the national collections

Group	No. of accessions in the collection	No. of distinct clones recognised
Beer	35	14
Musakala	26	8
Nakabululu	21	11
Nakitembe	32	13
Nfuuka	78	33
Total	**192**	**79**

6.4.5 Qualitative characters distinguishing accessions which are so similar but not identical

A number of accessions had values of their correlation and distance coefficients within the range of the ramets. They differed in one or two qualitative characters and so were assigned to different clones. Table 6.16 gives the frequency of occurrence the characters which distinguished these clones.

Plant stature, bunch shape and fruit orientation, fruit apices and anthocyanin presence on the petioles, in that order, were the most frequent characters distinguishing

accessions which were so similar but not identical. For stature, the most common change was dwarfism. Simmonds (1966) had previously noted this mutation in acuminata clones. The most common difference in fruit apex was intermediate versus blunt. Blunt apex of fruits was mentioned by De Langhe (1964) as a common mutation in acuminata clones. Fruits which are blunt can be reliably scored, and most fruits with blunt apices are slightly inflated as well. Bunch shapes tend to be influenced by the number of hands in the bunch and at times by the way fruits are arranged in the bunch. The growing conditions influence the number of hands in the bunch. Accessions which apparently differ in the qualitative character of bunch shape could be members of the same clone like members of Musakala clone set (C1, C2) and Nfuuka clone set (C17, C31). Studies on molecular markers would further confirm this.

Table 6.16: Frequency of occurrence of one or two characters separating pairs of accessions with correlation and distance coefficients within the range of known ramets in the different clone sets

Character and character states	Character change and frequency					
	Beer	**Musakala**	**Nakabululu**	**Nakitembe**	**Nfuuka**	**Total**
Stature	small/			small/		
-small	tall(1)			tall(1)		2
-semi-dwarf						
-tall	semidwarf/	semidwarf/			semidwarf/	
	tall(1)	tall(1)			tall(2)	4
Pseudostem blotching			absent/present			1
-extensive dark colouring of pseudostem (absent/present)			(1)			
Petiole anthocyanin	absent/		absent/			3
-absent/present	present (2)		present (1)			
-confined to margins						
-ventral side						
Bunch orientation	oblique/				oblique/	2
-subhorizontal	pendulous(1)				pendulous(1)	
-oblique						
-pendulous						
Bunch shape	rounded/					2
-rounded	rectangular(2)					
-rectangular	rectangular/				rectangular	2
-cylindrical	cylindrical(1)	rectangular/truncate(1)			/cylindrical (1)	1
-truncated						
Fruit orientation	no geotropic	no geotropic				3
-negatively	reaction/	reaction/				
geotropic	less recurved (1)		less recurved(2)			
-strongly recurved						
-less strongly		strongly/				2
recurved		less recurved(2)				
-no geotropic reaction						

Table 6.16 (Cont'd.)

Character and character states	Character change and frequency					
	Beer	**Musakala**	**Nakabululu**	**Nakitembe**	**Nfuuka**	**Total**
Fruit skin -variegation -green -glossy green					variegated/green (1)	1
Ovules -absent -present			absent/present (1)			1
Pulp colour before maturity -white -cream -orange brown					white/cream (1)	1
Pulp colour after maturity -white -cream -orange brown					cream/orange brown (1)	1
Fruit apices -blunt intermediate (1) -intermediate -bottle necked	blunt/ intermediate (2)		blunt/	blunt/ intermediate (1)		4
Persistent style -absence -present					absence/presence of style (2)	2
Fruit shape -rounded -rectangular -triangular -gourd shaped -slender	slender/triangular (1)				rounded/gourd shaped (1)	1 1
Rachis orientation -subhorizontal -oblique -pendulous	oblique/pendulous(1)					1
Persistent neuter flowers on rachis			semipersistent/ persistent (1)		semipersistent/ persistent (1)	3
-nude						
-semi-persistent						
-persistent						
Totals	**13**	**6**	**5**	**2**	**12**	**1**

Results in the table further suggest that Beer could be the clone set where mutations are most frequent followed by Nfuuka. Results also suggest that mutations are rare in Nakitembe. These can be interesting results in that Nfuuka was found to be the most heterogeneous clone set and it would not be surprising if it is the clone set with the most frequent occurrences of mutations.

6.4.5 Discussions

Multivariate methods are able to detect duplicates, i.e identify accessions which are phenetically similar. The results suggest that many accessions of Highland bananas are ramets of a single clone although they have different names. There are two principal reasons why accessions which represent the same clone may have different names. One reason is that new clones may be introduced in an area but without their local names. Farmers then give the introduced clone a name close to a clone that they know with similar characteristics. When they find out how different they are, they probably assign the introduced clone a new name. Another major reason why similar clones have different names is that they are grown in different banana regions by farmers speaking different dialects.

Although names of clones may differ in different regions, on many occasions the names have similar meanings. For example Namadhi, Nalusi, Nametsi and Enywamizi all mean water and are names used for a particular beer clone with abundant sap. A number of clones however receive new names because they have mutated in different localities, as observed by Shepherd (1957).

Some clones however, retain their original names in almost all the banana growing regions. For example Nakabululu, Embururu, Imbululu from three different regions share a similar name and are a single clone. Another example is Nalukira and Enyarukira. These clones which retain their names like Nakabululu, are widely used and do not show any observable change in most of the regions. Hence the name persists. In fact the name Nakabululu means short and compact but it also means to ripen at once. These characters are displayed in all the regions and are quite observable. When cooked, the pulp of the clone Nakabululu is also known in most of the Highland banana regions of Uganda to be tasty with a characteristic flavour.

Duplicates are however a problem in germplasm collections particularly those of clonal crops. They are collected and conserved because they had different names or they looked different because of phenotypic plasticity. All too often materials have simply been collected and maintained in germplasm collections without characterisation and hence duplicates accumulate causing problems of maintenance.

There is need to reduce duplication in the banana national collections because it is expensive to maintain them and management of the collections becomes impossible. However, precautions are necessary in the way they have to be reduced. Critical examination and studies are necessary to confirm that they are also genotypic duplicates. In the next section, views and proposals of reducing germplasm collections to a manageable size are discussed.

6.5 Part 2: Proposal for the establishment of a core collection of Ugandan Highland bananas

Lack of germplasm characterisation and evaluation, inadequate documentation and information about accessions, lack of fast, efficient and cost effective methods of germplasm screening are common problems encountered in most national germplasm collections (Morales *et al.*, 1995). These arise because the collections are large and unmanageable. Establishment of core collections would reduce most of these problems because it would be easier to work through smaller and better organised collections.

Estimates of the number of distinct clones have clearly indicated that there is considerable duplication within and between the National banana germplasm collections. It is therefore possible to develop a core collection which would be smaller but represent adequately the diversity in the two collections. Greater use of the East African Highland banana accessions could be made if a smaller number of accessions were selected, maintained and studied effectively. The objective of this section is to assess whether multivariate methods can assist in designating a core collection.

Different approaches to designating core collections have been discussed at length by various authors (Brown, 1989, 1995; Crossa *et al.*, 1995; Rao & Rao, 1995; Hamon *et al.*, 1995 and Cordeiro *et al.*, 1995). They all indicated the urgency and need for structured germplasm collections which could allow breeders, curators, students of biodiversity and various users to have easy access to germplasm collections.

Passport and characterisation data on accessions in a collection must be reasonably complete before a core collection can be established. Brown (1995) recommended that these data then be used for classification of the accessions into groups, often by hierarchical methods such as cluster analysis. He suggested the following data to be used; taxonomic data first (i.e morphology) then geographical origin, ecological origin, genetic markers and agronomic data in that order.

Crossa *et al.*, (1995) used multivariate methods based on morpho-agronomic data to develop a core collection of accessions of maize. 175 accessions initially selected on the basis of lodging and adaptation were evaluated for good agronomic characters using both cluster and principal component analysis in a stepwise manner. The accessions were of 3 types, those from the wet, the dry and the mixed ecologies. At each step of the analysis, a selection of half of the accessions within each group was included in the next step for further analysis until 27% of the original collection was left. Rao and Rao (1995) used geographical, taxonomic and agronomic data to divide a collection of sorghum into related groups from which representative samples were taken to form the core collection.

Galwey (1995) suggested that after the collection has been divided into groups, selection of accessions from groups could be done by either a constant strategy whereby equal numbers of accessions from each group are selected, or by a logarithmic strategy whereby the number of selected accessions is proportional to the logarithm of the number in the group, or by a proportional strategy whereby the number of selected accessions is proportional to the number in the group. Yonazawa *et al.* (1995) proposed in addition a random strategy whereby the entries are sampled from the collection randomly and the genetic diversity dependent strategy whereby the entries are sampled in proportion to the amount of genetic diversity in the groups. Yonezawa *et al.* (1995)

compared the five stratification strategies using both real and hypothetical collections. The genetic diversity dependent strategy was considered the best when the diversity in the collection is known in advance, otherwise the proportional strategy was considered optimal. Yonezawa *et al.* (1995) proposed further the use of a uniqueness value of accessions which measures the degree of genetic remoteness from other accessions. This is a modification of the proportional strategy and it involves calculating the genetic distances among the accessions from the topology of the phenograms in a stepwise manner.

Clone sets which were produced through multivariate analysis were used. Initially the aim was to reduce duplication and produce a reference collection to allow further testing of clone sets produced and find out how reliable they were. Besides reducing duplication, it was desirable to retain as much as possible the diversity found in the clone sets. In this study a core collection is proposed using taxonomic and geographical information. The 192 accessions of the East African Highland bananas used in this study were originally collected from three geographical locations within Uganda (Table 3.1, chapter 3). 89 accessions were from the central region (less than 1300 meters above sea level), 54 from the south-western region (above 1400 m) and 49 from the eastern region (above 1400 m). The accessions were divided into 5 clone sets using multivariate analyses.

The number of accessions to be retained is an important issue to be addressed when forming a core collection and it poses a major problem. The question is how large a fraction of the whole collection should be included in order to represent the diversity of the larger collection in the core collection. Brown (1989) recommended that the number of accessions should be about 10% of the original collection. He based his proposal on the sampling theory for selectively neutral alleles that had been put forward by Ewens (1972: quoted in Brown 1995). Yonezawa *et al.* (1995) mentioned that Ewens' model may not always hold in actual collections, let alone for the different species being maintained, the major reason being that it was based on the population not on accessions in a collection which are separated from the rest of the population and have been in isolation for some time. He then suggested his own model based on the calculation of the amount of genetic diversity found in the germplasm collection. Most curators are people interested in diversity and its management and it is easier for them to apply Brown's sample size of 10% than to make their own calculation on the basis of the model of Yonezawa *et al.* (1995).

There is no single figure (of the optimum sample size) one can apply to all collections since they range widely in scope, function and probably specificity.

If the 10% sample size of Brown (1995) was to be applied in our case, it would amount to 20 accessions. However, the 20 accessions could not sufficiently cover the diversity in the collection. Besides, the guidelines for optimal sample size of the germplasm collections suggested by Brown (1995) and Yonezawa *et al.*(1995), could not be clearly applied to our case because at present we have no information on allelic diversity of the crop and therefore cannot carry out the calculations done by Brown or Yonezawa *et al.* (1995). The information available to us is the morphological diversity of the clones and the different regions where the clones were collected from. The description of how core accessions were selected is therefore a representation of the morphologically diverse clones within each clone set and clones from the different banana growing regions of Uganda. 54 clones have been selected as explained below.

In describing the procedure which was carried out to select the accessions of the core collection, we shall adopt the nonconventional way of reading the hierarchy of the

phenogram upwards starting with the accessions as individual points which are grouped on an individual branch. A phenon line was drawn across each phenogram resulting from the cluster analyses of the 192 accessions from the two collections at a level corresponding to the extreme values of ranges that were obtained for known ramets (Figs 6.1, 6.2) and these were 0.759 for the correlation coefficient and 0.147 for the distance coefficient (Table 6.2). Accessions clustering above these levels were so similar that they may be the same clone so that they could be represented by just one accession. The majority of accessions which clustered at the first level of fusion above the phenon line belonged to the same clone. One of them would be retained. Then on that same branch or cluster in case the branch has more accessions, another accession would be selected if it was more diverse than the rest (i.e its node joining the rest at a higher distance or lower correlation). More than one accession from a subcluster above the phenon line would be retained if the accession came from different regions of the country. For example C6 and K142 (Figs. 6.1, 6.2) come from different localities and they were both retained although representing a single clone. A few clones were selected because they are prominent in the banana market and others because they are historically unique. C6 is a major commercial clone and K143 is considered one of the oldest clones, and it thrives in one area and it disappears and then it gradually reappears somewhere else. Table 6.17 indicates how selection was done in relation to the two phenograms. This first round of selection formed a reference collection (Table 6.17). Table 6.18 shows the percentage representation of each clone set and each region in the original collections and in the reference collection.

Almost half the accessions of the Highland bananas in the two collections were phenotypic duplicates on the basis of multivariate analyses of morphological characters. The question arises whether the duplicates should be discarded, now that the reference collection has been proposed. The answer is not as yet.

The reference collection is about half the size of the original collections. Duplication has been reduced but there is still much redundancy (repeated genotypes). The 102 reference accessions were advanced to the next stage of selection. A core collection was selected from the reference collection. Firstly, the accession with the name representing the clone set was selected. These were Namadhi (CB53) for Beer, Musakala (C2), Nakabululu (K47), Nakitembe (C19) and Nfuuka(K31).

The second round of selection was done in steps by selecting from each subcluster an accession more diverse than others. 54 accessions were retained and these appeared in the selection from each phenogram. The accessions in each clone set and from each region were not evenly distributed. While the aim was to select accessions with the highest diversity, it was also in order to represent all the regions. Table 6.19 gives both the list of accessions in the reference and core collection while Table 6.20 shows the percentage representation of each clone set and each region in the core collection.

Now that the core collection has been proposed, the next stage after setting it up is to prioritise research to be conducted using the core collection. These would be for example; improving methods of management and characterisation using various methods. Plant breeders and most other users are always interested in having a manageable number of genotypes that possess or are likely to possess the characters needed in their work. The core collection being in place would encourage greater use by the breeders themselves.

Figure 6.1: Phenogram showing group average clustering of matrix of correlation coefficients between 192 accessions in the Kawanda and Kabanyolo banana germplasm collections, with a phenon line marking the lowest correlation between known ramets. (C = accessions from Kawanda; K = accessions from Kabanyolo).

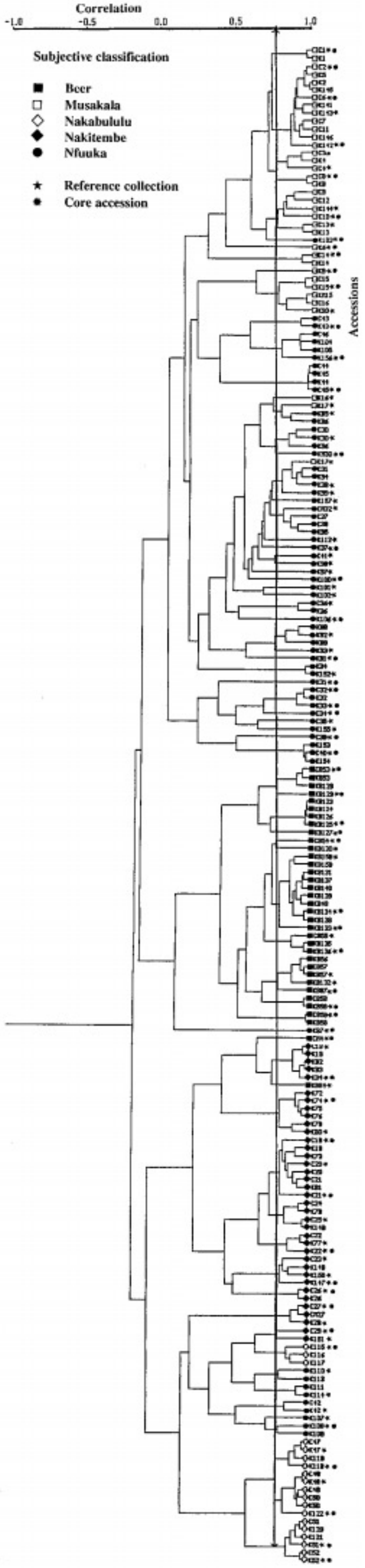


Table 6.17: Accessions for inclusion in the proposed reference collection. The order in the table follows that of the two phenograms to show the selection procedure.

Clone set	Selection of one from a subcluster	Reference collection
Musakala		
	C1/K1	C1
	C2-K142 (7 access.)	C2, C6, K142, K143
	C3, K4	C3
	C4	C4
	C8/K8	C8
	C9-C10 (4 access.)	C10, K144
	C13/K13	C13
	C14/K14	C14
Nfuuka		
	K9	K9
	K103	K103
	K6	K6
	C15/K15	K15
	KM15-K90 (3 access.)	K90
	C43/K43	K43
	C46-K156 (4 access.)	K156
	C44-C45 (4 access.)	C45
	K16	K16
	K17	K17
	K85/K86	K85
	K30-K96 (3 access.)	K30
	KM30	KM30
	C17	C17
	C31-K99 (4 access.)	K38, K99
	K157	K157
	CM32-K95 (4 access.)	CM32
	K112	K112
	K37	K37
	C41	C41
	K98	K98
	K97	K97
	K100	K100
	K101	K101
	K102	K102
	C36/K36	C36
	K106	K106
	K88-K89 (3 access.)	K92
	K93	K93
	K91	K91
	K34/K152	K152
	K31	K31
	C32-K33 (3 access.)	C32-K33
	C34	C34
	C35	C35
	K155	K155
	C39	C39
	K153-K154 (3 access.)	C40

Table 6.17 (Cont'd.)

Clone set	Selection of one from a subcluster	Reference collection
Beer		
	CB53-KB128 (3 access.)	CB53
	KB129	KB129
	KB123-KB125 (4 access.)	KB125
	KB127	KB127
	CB54	CB54
	KB130	KB130
	KB158-KB133 (10 access.)	KB133, KB134 KB158
	CB55-KB136 (3 access.)	CB55, KB136
	KB56-KB132	KB57, KB132
	CKB87	KB87
	CB59/KB59	KB59
	CB58/KB58	CB58
Nfuuka		
	K87	K87
Beer		
	CM4	CM4
Nakitembe		
	C18/K18	C18
	K82-K84 (3 access.)	K84
Beer		
	KB84	KB84
Nakitembe		
	K72-K76 (4 access.)	K74
	K79, K80	K80
	C19-K81 (7 access.)	C19, C20
	K21	K21
	C24-K149 (4 access.)	C25
	C22/K27	K77
	K22	K22
	C23-K150 (3 access.)	C23, K150
	K147	K147
	C26/K26	C26
Nakabululu		
	C27-K28 (3 access.)	C27, K28
	C29	C29
	K151	K151
Nfuuka		
	K115-K117 (3 access.)	K115
	K110, K113	K110
	K111, K114	K114
	C42/K42	K42
	K107	K107
	K108/K109	K108
Nakabululu		
	C47-K118 (4 access.)	K47, K118
	C48-K50 (5 access.)	K48, K50
	K122	K122
	C51-K51 (4 access.)	K51
	C52/K52	K52

Table 6.18: Comparison of the proposed reference collection with the original collection as regards proportion of accessions from the different clone sets from the different regions.

Clone set	Original collections	Reference collection
Beer		
	35 (18%)	18 (18%)
	7 Central (4%)	4 Central (4%)
	17 South western (9%)	8 South western (8%)
	11 Eastern (6%)	6 Eastern (6%)
Musakala		
	26 (14%)	12 (12%)
	13 Central (7%)	5 Central (5%)
	8 South western (4%)	4 South western (4%)
	5 Eastern (3%)	3 Eastern (3%)
Nakabululu		
	21 (11%)	11 (11%)
	8 Central (4%)	4 Central(4%)
	6 South western (3%)	3 South western (3%)
	7 Eastern (4%)	4 Eastern (4%)
Nakitembe		
	32 (17%)	14 (14%)
	15 Central (8%)	7 Central (7%)
	8 South western (4%)	3 South western (3%)
	9 Eastern (5%)	4 Eastern (4%)
Nfuuka		
	78 (41%)	47 (46%)
	34 Central (18%)	21 Central (21%)
	26 South western (14%)	14 South western (14%)
	18 Eastern (9%)	12 Eastern (12%)
Total	**192**	**102**

Setting up a core collection is the beginning of work which has not been going on because the size of the collection was big. As Pickersgill (1995) mentioned, many different characters besides morphology should be used during evaluation in order to detect duplicates and distinguish between similar phenotypes which have evolved through convergence. She recommended molecular markers as very useful in detecting possible duplicates. Galwey (1995) also recommended surveys of both the core and the whole collection using biochemical or molecular markers to establish whether there are more significant pockets of variation that have escaped inclusion in the core.

Figure 6.2: Phenogram showing group average clustering of matrix of distance coefficients between 192 accessions in the Kawanda and Kabanyolo banana germplasm collections, with a phenon line marking the maximum distance between known ramets (C = accessions from Kawanda; K = accessions from Kabanyolo.

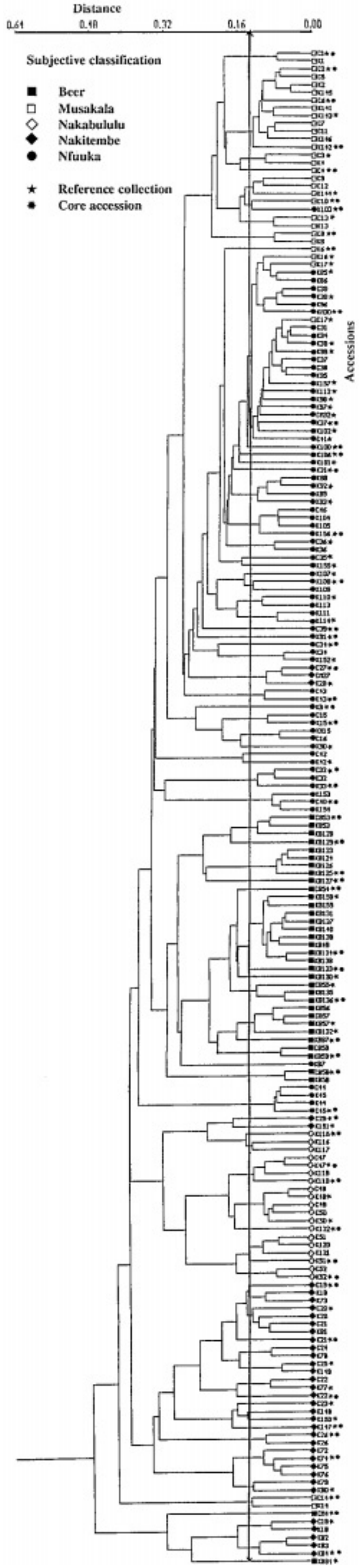


Table 6.19: Core collection selected from the reference collection. * Accessions in the core collection

Code number	Accession
Beer clone set	
CM4	Mudwale Beer (E)*
CB53	Namadhi (E)*
CB54	Mende (C)*
CB55	Bagandeseza (C)
KB57	Nalukira (C)
CB58	Endirira (W)*
KB59	Katalibwambuzi (W)*
KB84	Oruhuuna Beer (W)
KB87	Namunyere Beer (E)*
KB125	Entanga (W)*
KB127	Bwara (W)*
KB129	Nametsi (E)*
KB130	Nalusi E)
KB132	Shombo-obureku (W)
KB133	Kibagampera (W)*
KB134	Engumba (C)*
KB136	Engambani (W)*
KB158	Mwanga (W)
Musakala clone set	
C1	Muvubo (C)*
C2	Musakala (C)*
C3	Nakibizzi (C)
C4	Mudwale Cooking (E)*
C6	Mayovu (E)*
C8	Mukazialanda (W)*
C10	Muturit (E)*
C13	Wansimirahi (W)
C14	Namunwe (C)*
K142	Rwamigongo (W)*
K143	Lwewunzika (C)
K144	Mujuba (W)
Nakabululu clone set	
C27	Nakyetengu (C)*
K28	Ekitetengwa (W)
C29	Kibuzi (C)*
K47	Nakabululu (C)*
K48	Nakasabira (E)
K50	Kazirakwe (W)
K51	Butobe (C)*
K52	Kaitabunyonyi (E)*
K118	Bifusi (E)*
K122	Mukite (E)*
K151	Salalugazi
Nakitembe clone set	
C18	Namaliga (C)
C19	Nakitembe (C)*
C20	Nakitembe-Nakamali (C)
K21	Namulondo (C)*
K22	Nakitembe-Nakawere (C)*
C23	Nakitembe-Omunyoro (W)
C25	Nakibuule (C)
C26	Mbwazirume (C)*
K74	Enjagata (W)*
K77	Luvuta (E)

Code number	Accession
Nakitembe clone set (Cont'd.)	
K80	Nasaala (E)
K84	Oruhuuna cooking (W)*
K147	Lisindaalo (E)*
K150	Bikowekowe (E)
Nfuuka clone set	
K6	Mayovu E)*
K9	Nalugolima (C)*
K15	Siira (C)*
K16	Atwalira (C)
C17	Kabucuragye (W)
K17	Kabucuragye (W)
K30	Nakinyika (C)
KM30	Enyakinika (W)*
K31	Nfuuka (C)*
C32	Namwezi (C)*
CM32	Namwezi Black (C)
K33	Kulwoni (E)*
C34	Ndyabalangira (C)*
C35	Tuula-twogere (C)
K36	Nakabinyi (C)
C37	Enyeru (W)*
K38	Nakhaki (E)
C39	Tereza (C)*
C40	Nakawere (C)*
C41	Enjeriandet (E)
K42	Nante (E)
K43	Bitambi (C)*
C45	Entukura (W)*
K85	Entazinduka (W)
K87	Namunyere Cooking (E)*
K90	Nassaba (E)
K91	Rwezinga (W)
K92	Mutta-Ngendo (C)
K93	Nabusa (C)
K97	Rwasha (W)
K98	Sitakange (C)
K99	Serunjogi (C)
K100	Namafura (E)*
K101	Nasuuna (C)
K102	Kabende (E)
K103	Enyamanshari (W)*
K106	Mukaddealikisa (C)*
K107	Lusumba (C)
K108	Namamuka (E)*
K110	Enyabakazi (W)
K112	Khabushi (E)
K114	Namakhumbu (E)
K115	Bukumo (W)*
K152	Enzirabushera (W)
K155	Kaasa (W)
K156	Kibalawo (C)*
K157	Enyaruyonga (W)

Table 6.20: Comparison of the proposed core collection with the reference collection as regards proportion of accessions from the different clone sets from different regions.

Clone set	Reference collection	Core collection
Beer	18 (18%)	12 (22%)
	4 Central (4%)	2 Central (4%)
	8 South western (8%)	5 South western (9%)
	6 Eastern (6%)	5 Eastern (9%)
Musakala	12 (12%)	8 (15%)
	5 Central (5%)	3 Central (6%)
	4 South western (4%)	2 South western (4%)
	3 Eastern (3%)	3 Eastern (6%)
Nakabululu	11 (11%)	7 (13%)
	4 Central (4%)	3 Central (6%)
	3 South western (3%)	1 South western (2%)
	4 Eastern (4%)	3 Eastern (6%)
Nakitembe	14 (14%)	7 (13%)
	7 Central (7%)	4 Central (7%)
	3 South western (3%)	1 South western (2%)
	4 Eastern (4%)	2 Eastern (4%)
Nfuuka	47 (40%)	20 (37%)
	21 Central (21%)	10 Central (19%)
	4 South western (14%)	5 South western (9%)
	2 Eastern (12%)	5 Eastern (9%)
Total	**102**	**54**

It is therefore recommended that a core collection of Highland bananas be established in the field at two locations, one in the central lowland and another one in the highlands. Setting up a core collection in two different zones allows expression of characters among clones in the two environments to confirm further those clones that are true phenotypic duplicates and those which may not. Further advanced studies will also be carried out on the core collection while the main collection and the rest of the reference collection not included in the core collection are gradually being transferred to *in vitro* preservation.

Chapter 7

STUDIES ON BANANA CLONES IN FARMERS' FIELDS

Studies on the national banana collections have shown that different growing conditions in two similar ecological locations did not prevent the classification of the East African Highland bananas into identifiable groups. The studies did, however, indicate that many accessions in the national banana collections were similar phenotypically and the number of distinct clones in these collections was limited. At the end of the study, a revised classification based on the results of these studies and a reference collection and core collection were proposed.

The classification proposed is supposed to allow for further modification as more accessions are incorporated. The next step was to test this classification to see whether clones growing in different environments can be placed in their appropriate clone sets. Shepherd (1957) suggested that individual clones can change their phenotypes when they are grown in different environments and that a workable classification would not be easy to construct. The following study therefore was an attempt to see whether farmers' clones can be placed accurately into their appropriate clone sets in spite of growing under different management systems, and also to assess whether there are possible new clones not represented in the collections.

The clones of East African Highland bananas have been selected and distributed by farmers themselves. Cultivation has been largely in their hands and many historical changes in the crop have been observed by them. Farmers' knowledge of the crop will also form important links to the classification of the crop (van der Maesen, 1988).

This phase of study on farmers' clones therefore involved a morphometric study which was conducted on clones selected from the three major growing zones of bananas in the country. Cluster analyses and discriminant analyses were carried out to compare farmers' accessions with the accessions in the reference collection.

7.1 Multivariate analysis of farmers' clones together with the reference collection

7.1.1 Materials and methods

7.1.1.1 Site selection

Farmers' banana fields which were under study were located in the eastern highlands (the Elgons), the central lowlands and the south-western highlands. The central lowlands are located in the Banana-Robusta coffee agricultural system while the

western and eastern highlands are located in the Montane agricultural system (Fig.7.1). Eight sites were selected. Six of the sites were those formerly used by the National banana programme diagnostic survey which took place in 1993/94 to assess and quantify farmers' problems in relation to production of bananas in Uganda (Gold et al., 1993) (appendix 2). Two other sites (sites 2 and site 6) were selected. They are regions which have had a recent influx of immigrants. Immigrants would bring new clones into the area or make a change in the food habits of people in the area.

7.1.2 Sites in the Banana-Robusta coffee agricultural system

Study sites in this system were located in Luwero district, Semuto subcounty (site 6) and Rakai district, Kagamba subcounty (site 5) (see Fig. 7.1). The areas under study lie 1080-1330 metres above sea level (Table 7.1). They are relatively wet throughout the year. However, some parts of Kagamba (site 5) are dry due to being in the rain shadow of the highlands, while Semuto (site 6) has two dry months in August and September when the second rains are uncertain. The temperatures are high with mean annual maximum temperatures between 22.5oC and 27.5oC. The soils are sandy loams and sandy clay loams with a pH range from 5.5-6.4. Formerly the agricultural systems in Semuto (site 6) were banana-based but due to declining soil fertility, weevils, nematodes and diseases, banana production declined (Gold et al., 1993). However, banana plantations are being re-established.

7.1.3 Sites in the Montane system

Banana fields in the Montane system are located either along the slopes or on the foothills of the mountains. Study sites in the eastern highlands were located in Mbale, Butiru and Buginyanya subcounties (site 7) and Kapchorwa, Kaseren subcounty (site 8). The south western highland sites were Mbarara, Rugaga subcounty (site 4), Bushenyi, Ryeru subcounty (site 3), Kabale, Bukinda subcounty (site 1), and Kabarole, Bukuku subcounty (site 2) (Fig. 7.1). All these sites are high altitude areas ranging from 1299-1870 metres and thus they are mostly cooler than the surrounding areas during both the rainy and dry periods. The mean annual temperatures are 25oC. The climate is highly influenced by the location of these sites because the higher the location, the lower the temperatures. This means that the maturation period (shooting to harvest) of the crop will be longer than the 5 months which is commonly the maximum period in the banana-coffee zone due to lower temperatures. Rainfall is bimodal with peaks in March/April and August/October, and a total which ranges from 1000 millimetres to 1450 millimetres per annum. The soils are generally deep, well structured and fertile. Most are dark to brown volcanic soils, rich in organic matter, but some are red sandy clay loams. Bananas now form by far the largest portion of the diet of people in both the south-western and eastern highlands, which was not the case in the 1950s.

7.1.4 Farm selection

10 farms were selected for study at each site although not all of them were used. The farms studied were those containing 50 or more banana mats which were in at least the second or third ratoon crops. The 50 mats criterion was set to ensure presence of a "banana system" and to provide adequate sample sizes.

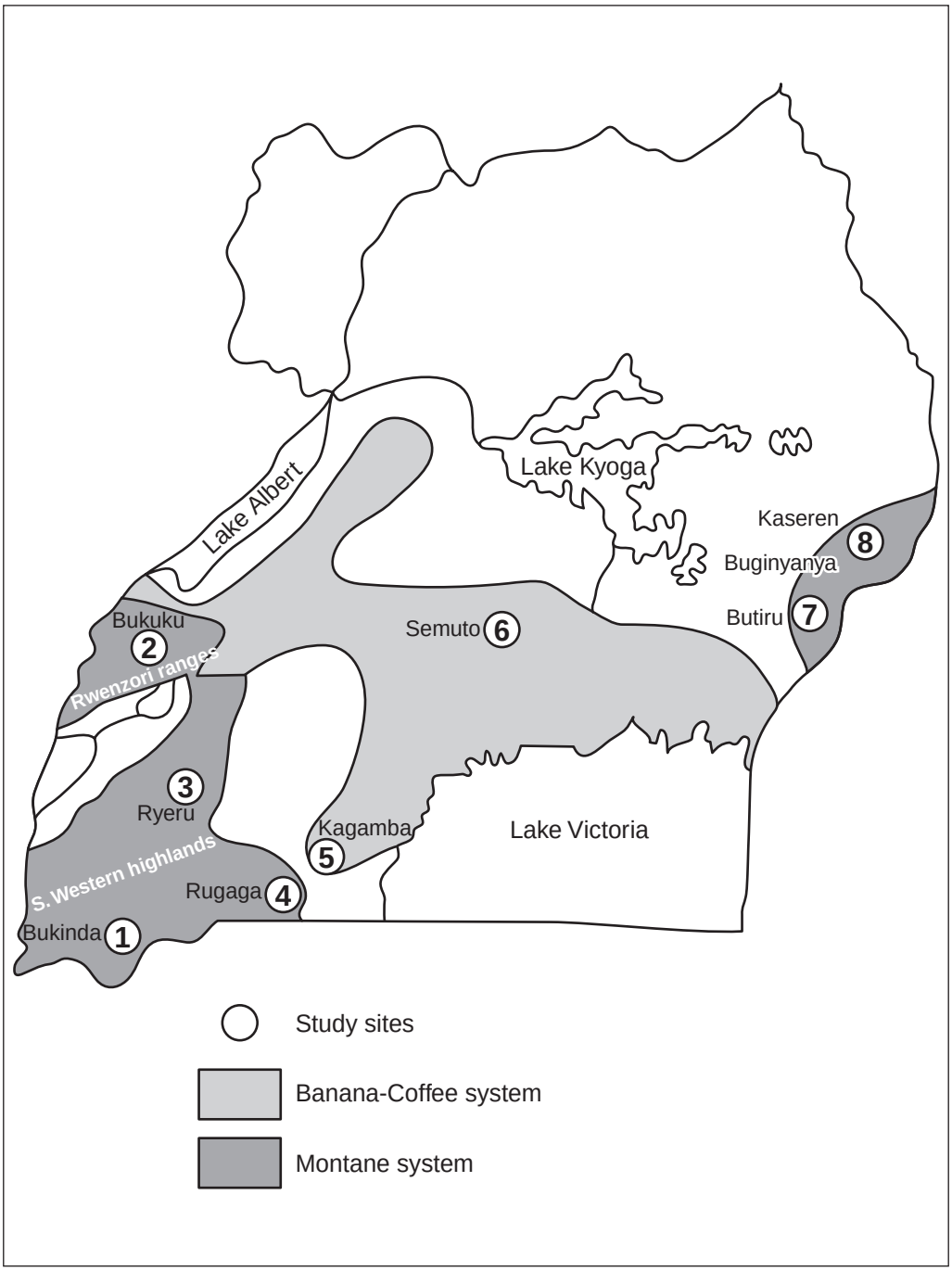


Figure 7.1: Study sites in the banana coffee agricultural system and the Montane agricultural system in Uganda.

Table 7.1: Agroecological conditions of the study sites

Agroecological conditions	SITES							
	Central region		South western region				Eastern region	
	Kagamba (site 5)	Semuto (site 6)	Bukinda (site 1)	Bukuku (site 2)	Ryeru (site 3)	Rugaga (site 4)	Butiru (site 7)	Kaseren (site 8)
Altitude (m)	1190-1330	1082-1250	1760-1830	1520-1560	1340-1420	1430-1830	1299-1524	1820-1870
Mean annual rainfall (mm)	1200	1000	1350	1000	1450	1200	1250	1400
Mean annual temperature (0°C)	25	25	17.5	19	25	22	22	18
Soils	sandy clay loam	sandy loam	reddish brown clays	volcanic	volcanic	red sandy loams	red sandy loams	volcanic
Mulching	sometimes	rare	year round	year round	year round	year round	most of the time	most of the time
Weeding	by hoe	by hoe	by hand	by hand	by hand	hand and hoe	hand and hoe	by hoe
Plant density (per stool)	3 plants	2-5 plants	3 plants	2-5 plants	3 plants	3 plants	3 plants	3-5 plants
Spacing	irregular	irregular	2.5 x 2.5m	irregular	3 x 3 m	3 x 3 m	3 x 3 m	irregular

7.1.5 Accession selection

On each farm, 25 stools were randomly selected and were named by the farmer. A clone with five healthy stools was then used to select 5 plants, one per stool. These were used for data taking. Accessions included in this study were those whose names were not included in the previous studies; those which had the same name as any clone in the collection, in order to check whether phenotypic variation overrides genetic differences; and those which had enough sample size on the selected farm. Farmers grow a complex mixture of Highland banana clones so that to get a complete set of five healthy stools was not easy.

Sixty accessions were initially selected from the farmers' field sites. Farmers customarily cut off male buds one to two months after shooting (chapter3). Many of them forgot about the marked plants and they removed the male buds and even harvested some bunches before data could be recorded. Removal of a bunch and male buds meant that more than half of the characters were lost. Cluster analysis in this phase therefore contains more missing data than the previous analyses.

Accessions for which many characters could not be scored were discarded from the analysis. 14 accessions therefore had to be discarded. 46 accessions from the farmers' fields were included in cluster analysis. 16 were from the central region, 19 were from the south western highlands and 11 were from the eastern region (Table 7.2).

Twenty of the 46 accessions from the farmers' fields had similar names to accessions in the collections. These were given numbers similar to those of the counterpart accessions in the collections. Other accessions were given unique numbers as their names were being met for the first time. All selected accessions were marked before taking data to assist both the farmers and the researcher to know the plants being used in the study.

One hundred and two accessions which represented the reference collection from the previous study were used in cluster analysis together with the 46 accessions from the farmers' fields to keep the phenogram a reasonable size (Table 7.3).

In discriminant analysis, however, 190 accessions previously classified in the study of multivariate analyses of the national banana collections were used as a training set. 9 accessions from the farmers' fields were excluded from discriminant analysis for having missing data (F30, F46, F47, F73, F104, F112, F166, FB179 and FB180). This means that 37 accessions from the farmers' field represented a hold-out set in this analysis.

7.1.6 Selection of characters and methods of analysis

The 61 characters used in the previous analyses were also used in this cluster analysis. The pairwise estimation of resemblances between accessions was carried out using the correlation coefficient. The average taxonomic distance coefficient was discarded because it emphasised differences in size more than the correlation coefficient. Cluster analysis of similarity was conducted using the group average clustering method.

In classificatory discriminant analysis however, 16 characters used in the previous discriminant analysis were used. The analysis was carried out using the k=5 nearest neighbour technique to classify the 37 accessions from the farmers' fields into the five previously recognised clone sets.

Table 7.2: 46 accessions studied in farmers' fields

Code number	Clone	Site	Region	Group
FB128	Enywamaizi	Bukuku (2)	South western	Beer
FB138	Engumba	Kagamba (5)	Central	Beer
FB176	Ensika	Bukinda (1)	South western	Beer
FB177	Engote	Bukinda (1)	South western	Beer
FB178	Entundu	Rugaga (4)	South western	Beer
FB179	Baleka	Bukuku (2)	South western	Beer
FB180	Omuzo	Ryeru (3)	South western	Beer
FB181	Nakanyala	Rugaga (4)	South western	Beer
F2	Musakala	Semuto (6)	Central	Musakala
F4	Murure	Butiru (7)	Eastern	Musakala
F7	Kisana	Semuto (6)	Central	Musakala
F143	Lwewunzika	Kagamba (5)	Central	Musakala
F163	Batule	Semuto (6)	Central	Musakala
F169	Enyoya	Rugaga (4)	South western	Musakala
F174	Namunget	Kaseren (8)	Eastern	Musakala
F47	Nakabululu	Butiru (7)	Eastern	Nakabululu
F115	Bukumo	Rugaga (4)	South western	Nakabululu
F165	Mukubakkonde	Kagamba (5)	Central	Nakabululu
F175	Mburiondet	Kaseren (8)	Eastern	Nakabululu
F73	Entaragaza	Ryeru (3)	South western	Nakitembe
F74	Enjagata	Ryeru (3)	South western	Nakitembe
F151	Salalugazi	Semuto (6)	Central	Nakitembe
F161	Nakitembe-Omusoga	Semuto (6)	Central	Nakitembe
F170	Kitika	Bukuku (2)	South western	Nakitembe
F171	Kongowet	Kaseren (8)	Eastern	Nakitembe
F172	Endyabawali	Bukuku (2)	South western	Nakitembe
F182	Nakitembut	Kaseren (8)	Eastern	Nakitembe
F30	Nakinyika	Semuto (6)	Central	Nfuuka
F31	Nfuuka	Semuto (6)	Central	Nfuuka
F32	Namwezi	Semuto (6)	Central	Nfuuka
F37	Enyeru	Ryeru (3)	South western	Nfuuka
F41	Enjeriandet	Kaseren (8)	Eastern	Nfuuka
F46	Namande	Semuto (6)	Central	Nfuuka
F100	Namafura	Butiru (7)	Eastern	Nfuuka
F104	Nambogo	Butiru (7)	Eastern	Nfuuka
F107	Lusumba	Semuto (6)	Central	Nfuuka
Fl10	Enyabakazi	Bukuku (2)	South western	Nfuuka
Fl12	Khabusi	Butiru (7)	Eastern	Nfuuka
F156	Kibalawo	Semuto (6)	Central	Nfuuka
F160	Katwalo	Semuto (6)	Central	Nfuuka
F162	Ntika	Semuto (6)	Central	Nfuuka
F164	Nakibira	Semuto (6)	Central	Nfuuka
F166	Nabusolo	Butiru	Eastern	Nfuuka
F167	Enshenyi	Bukinda (1)	South western	Nfuuka
F168	Enyambo	Rugaga (4)	South western	Nfuuka
F173	Rwambarara	Rugaga (4)	South western	Nfuuka

Table 7.3: Number of accessions from each subjective clone set represented in this analysis

Clone set	Reference collection	Farmers' fields	Total
Beer	16	8	24
Musakala	12	7	19
Nakabululu	7	4	11
Nakitembe	17	8	25
Nfuuka	50	19	66
Total	**102**	**46**	**148**

7.2 Results

7.2.1 Phenogram based on correlation coefficients

In the phenogram (Fig.7.2) resulting from the group average cluster analysis, the accessions formed clusters corresponding to clone sets similar to those in the previous analyses. The different conditions in farmers' fields did not make farmers' accessions form distinct clusters, separate from accessions in the reference collection. They were placed in the clone sets to which they had been subjectively assigned.

Ten of the 23 accessions from the farmers' fields which shared names with accessions already studied clustered first with their counterparts in the collections (Fig.7.2). Although 4 other accessions (F7, F46, F104, FB128) shared names with accessions in the collection, their counterparts from the collections were not included in this analysis. However, F7 clustered with K142, which represents the clone to which C7 belongs, while F46, F104 and F128 also joined clusters to which their counterparts would belong.

Some accessions were not placed where they were expected to be. C14, an accession which was inconsistently placed in previous cluster analyses, clustered with two field accessions (F169, F174) far away from Musakala cluster where they were expected to be. They formed their own subcluster.

F170 and F182 did not cluster with Nakitembe accessions but clustered with C27, K28 and C29 (Nakabululu accessions) at a low similarity level. A small discrete cluster of Nfuuka (K30, F30, KM30, K97) also clustered with Musakala but this could be because of the reduced number of accessions.

Cluster analysis imposes some form of structure on the data even if no real structure exists (Sokal & Rohlf, 1962; Sneath, 1976). In order to assess further the classification of farmers' accessions, a classificatory discriminant analysis was conducted. 9 accessions (F30, F46, F47, F73, F104, F112, F166, FB179 and FB180) were excluded from classificatory discriminant analysis because they had missing data. These accessions were satisfactorily placed by cluster analysis (Fig.7.2).

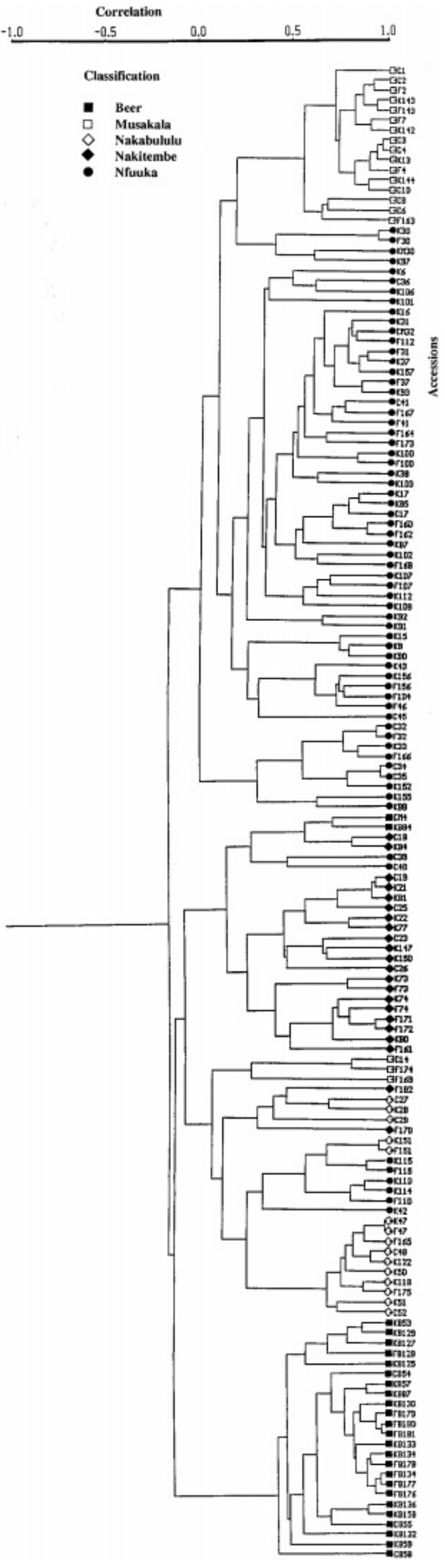
7.2.2 Classificatory discriminant analysis

Table 7.4 gives the classification of the 37 accessions. 34 accessions were classified into the clone sets to which they had been subjectively assigned with a probability of 100%. There was some doubt about the classification of the remaining 3 accessions. F74 which had been subjectively classified as Nakitembe had 67% probability of

Table 7.4: 37 accessions from farmers' fields as classified by classificatory discriminant analysis.

Accessions	Subjective classification	Cluster analysis	Classificatory discriminant analysis
FB128	Beer	Beer	Beer
FB134	Beer	Beer	Beer
FB176	Beer	Beer	Beer
FB177	Beer	Beer	Beer
FB178	Beer	Beer	Beer
FB181	Beer	Beer	Beer
F2	Musakala	Musakala	Musakala
F4	Musakala	Musakala	Musakala
F143	Musakala	Musakala	Musakala
F163	Musakala	Musakala	Musakala
F169	Musakala	C14, F169,F174	Musakala
F174	Musakala	C14, F169, F174	Musakala
F165	Nakabululu	Nakabululu	Nakabululu
F175	Nakabululu	Nakabululu	Nakabululu
F74	Nakitembe	Nakitembe	Nakitembe (67%) Musakala (33%)
F151	Nakitembe	K151, K115, F115, K110, F110, K42	Nakabululu
F161	Nakitembe	Nakitembe	Nakitembe
F170	Nakitembe	C27, K28, C29, F182	Nakabululu
F171	Nakitembe	Nakitembe	Nakitembe (83%) Musakala (17%)
F172	Nakitembe	Nakitembe	Nakitembe (83%) Musakala(17%)
F182	Nakitembe	C27, K28, C29, F170	Nakabululu
F31	Nfuuka	Nfuuka	Nfuuka
F32	Nfuuka	C32, K32, F166	Nfuuka
F37	Nfuuka	Nfuuka	Nfuuka
F41	Nfuuka	Nfuuka	Nfuuka
F100	Nfuuka	Nfuuka	Nfuuka
F104	Nfuuka	Nfuuka	Nfuuka
F107	Nfuuka	Nfuuka	Nfuuka
F110	Nfuuka	Nfuuka	Nfuuka
F115	Nfuuka	Nfuuka	Nfuuka
F156	Nfuuka	K43, C45, F46 F104, K156	Nfuuka
F160	Nfuuka	Nfuuka	Nfuuka
F162	Nfuuka	Nfuuka	Nfuuka
F164	Nfuuka	Nfuuka	Nfuuka
F167	Nfuuka	Nfuuka	Nfuuka
F168	Nfuuka	Nfuuka	Nfuuka
F173	Nfuuka	Nfuuka	Nfuuka

Figure 7.2: Phenogram derived from cluster analysis (UPGMA method) of correlation matrix involving 46 accessions from farmer's fields and 102 accessions in the reference collection.



belonging to the Nakitembe and 33% of belonging to Musakala. F171 and F172, both subjectively classified as Nakitembe, had each 83% probability of belonging to the Nakitembe clone set, and 17% of belonging to the Musakala clone set. 3 other accessions (F151, F170 and F182) had been subjectively classified as Nakitembe accessions but were placed in Nakabululu. The positions of these six accessions are briefly considered in the discussion section of this chapter. However, there was agreement between the subjective and phenetic classifications with regard to positions of the majority of the accessions.

Although classificatory discriminant analysis has placed correctly the majority of accessions in their appropriate groups, it cannot tell us how similar farmers' accessions were to each other and to those in the collection and hence cannot assess whether these accessions were the same as those already in the collections or were new clones not represented in the collection.

7.2.3 Assessment of whether accessions in farmers' fields belong to clones already represented in the collections

7.2.3.1 Accessions from the farmers' fields which shared names with accessions in the collections

Table 7.5 shows correlation coefficients between members of the 19 pairs of accessions which shared the same names and were both included in the analysis. 14 of the 19 pairs had high correlation values, which were within the range previously established for ramets. Members of each pair were identical in their qualitative characters and were thus considered to be members of the same clone. These pairs of accessions were F2/C2, F4/C4, F143/K143, F30/K30, F31/K31, F32/K32, F47/C47, FB134/KB134, F151/K151, F156/K156, F73/K73, F100/K100 F110/K110, F115/K115. Majority of these pairs of accessions had their field counterparts growing in the lowland areas. Results in the table show that there were differences in average correlation values between pairs having counterpart accessions growing in the highlands and those growing in the lowland. The pairs of accessions with counterparts growing in the highlands have generally lower average correlations than those previously established for ramets.

However, one pair (K107 and F107) with counterpart accession from the lowland areas was identical in all the qualitative characters but had slightly lower correlation values than those previously established for ramets. F107 (Lusumba) was studied at the Semuto site. It was not a vigorous clone. As recorded in section 7.1.2, the second rains are always uncertain in Semuto site and thus there is a prolonged dry season, hence plants become a bit stunted. K107 in Kabanyolo collection was also not vigorous. The growing conditions of K107 were not good since it was in the eroded block of Kabanyolo collection. K107 and F107 probably represented a single clone.

Table 7.5: Correlation coefficients between accessions from farmers' fields with the same name as an accession in the collection.

Accessions from the lowlands	Correlation coefficient	Accessions from the highlands	Correlation coefficient
F2, C2	0.915	F4,K4	0.913
F143, K143	0.933	F37,K37	0.639
F30, K30	0.942	F41, C41	0.651
F31, K31	0.891	F73, K73	0.793
F32, K32	0.949	F74, K74	0.652
F47, C47	0.988	F100, K100	0.840
F107, K107	0.706	F110, K110	0.853
FB134, KB134	0.932	F112, K112	0.600
F151, K151	0.956	F115, K115	0.893
F156, K156	0.768		
Average similarity	**0.900**		**0.759**
Range of values for known ramets	**0.759-0.988**		

The other 4 pairs of accessions which shared the same name were less similar than the known ramets (Table 7.6). These pairs were F37/K37 (Enyeru) and F74/K74 (Enjagata) from the south western highland, F41/C41 (Enjeriandet) and F112/K112 (Khabusi) from the east. All are major commercial clones in the south western and the eastern highland regions. These 4 pairs of accessions belong to clone sets Nfuuka and Nakitembe.

Nfuuka clone set

F37 and K37 (Enyeru), were each more similar to CM32 (Namwezi Black) and K93 (Nabusa) than to each other (Table 7.6). In the previous chapter, C37, K37, CM32 and K93 were shown to be probably members of a single clone. The similarity between F37 and CM32 is therefore not surprising. F37 was identical to K37 and CM32 in all qualitative characters and is therefore considered to belong to this clone also. Differences were in the quantitative characters which were attributed to different growing conditions.

K37 was also similar to three other accessions in farmers' fields. These were F31 (Nfuuka from the central region), F167 (Enshyenyi from the south west) and F168 (Enyambo from the south west). These accessions differed from K37 in all 13 quantitative characters but were identical to K37, and to each other, in all their qualitative characters and therefore likely to belong to one clone. Enyambo (F168) has been introduced from Tanzania. It generally grows at lower altitude (below 1300 meters) than Enshenyi. However, F168 was studied at Rugaga (site 4) at an altitude of 1430-1470 meters. Farmers in Rugaga found Enyambo did not perform as well as in Tanzania (Bukoba region), probably due to differences in altitude.

Table 7.6: Accessions from farmers' fields which were more similar to other accessions than to their counterparts in the collections.

Correlation coefficients between accessions which shared the same name (figures in parentheses show differences in quantitative/qualitative characters)	Correlation with other accessions	Differences in quantitative/ qualitative characters	Conclusion
F37/K37 = 0.639 (13/0)	F37/CM32 = 0.799	13/0	All probably represent a single clone
	K37/CM32 = 0.805	12/0	
	F37/K93 = 0.861	13/0	
	K37/F31 = 0.891	13/0	
	K37/F167 = 0.817	13/0	
	K37/F168 = 0.786	13/0	
	F37/F167 = 0.774	12/0	
	F37/F168 = 0.759	13/0	
F41/C41 = 0.651 (13/0)	C41/F167 = 0.773	12/0	Single clone
F112/K112 = 0.600(12/0)	F112/K31 = 0.800	13/0	All probably represent a single clone
	F112/F31 = 0.853	13/0	
	F112/CM32 = 0.832	13/0	
	F112/K37 = 0.772		
F74/K74 = 0.652 (13/0)	K74/F171 = 0.800	13/0	Single clone
	F74/F151 = 0.853	12/0	
	K74/F172 = 0.807	13/0	
	F74/F172 = 0.834	13/0	

F112 (Khabusi) was more similar to K31, F31, K37 and CM32 (these accessions, together with K112, were all regarded as a single clone in the previous chapter) than to K112. We take it that these are probably all the same clone. However, Khabusi is a clone well adapted to the eastern highlands and was bound to show differences with its counterpart accession in the Kabanyolo ollection.

Farmers in the field mentioned that K112/F112 was the same clone as C29 (Kibuzi from the central region). C29 has pendulous compact bunches, inflated blunt fruits and semi-persistent floral parts along the rachis, but K112/F112 has oblique compact bunches with rectangular non inflated fruits with intermediate shaped apices and hence was considered different from C29.

C41, an accession regarded as belonging to the same clone as the other Nfuuka accessions discussed in this section, differed in quantitative characters from F41. Bananas at Kaseren (site 8), where F41 was growing, were not well maintained.

Since F41 was more similar to C41 (Table 7.6) than to any other accession in the analysis and the two were identical in the qualitative characters that were scored. F41 also belongs to the clone Enyeru.

Nakitembe clone set

F74 and K74 were each more similar to F171 (Kongowet from the east) and F172 (Endyabawali from the west) than to each other. Kongowet and Endyabawali were identical to one another and also identical to F74 and K74 in all their qualitative characters. They belong to the same clone as F74 and K74. Differences between K74

and F74 were due to growing conditions. F74 is a commercial clone of the western region well maintained in all farms. It was bound to be different from K74 which was in the eroded block at Kabanyolo. F74 is grown in the highlands and well adapted there. F171 and F172 are also from high altitude zones. Higher altitude may have a part to play in the differences between F74 and K74. Some clones have been considered to be adapted to highland ecologies (Karamura *et al.* 1996) and F74 may be one of them.

Four accessions (F7, F46, F104 and FB128) from the farmers' fields shared names with accessions in the collections but their counterpart accessions were not included in the analysis. They are however compared with accessions representing the clones to which their counterparts belong (Table 7.7).

F7 (Kisansa from central region) was more similar to K142 than to any other accession in the analysis. F7 was also identical in qualitative characters to K142. These results suggest that F7 belongs to the same clone as K142 and hence as C7.

Table 7.7 Accessions from farmers' fields which shared names with accessions in the collections but not included in this analysis.

Most similar accession in the analysis	Accession in the analysis most similar to this	Quantitative/ qualitative character differences	Conclusion
F7	K142 = 0.900	(13/0)	Same clone
F46	F104 = 0.760	(13/0)	Same clone
F128	KB53 = 0.772	(13/0)	Same clone

F46 (Namande from the central region) and F104 (Nambokho from the eastern region) were more similar to each other than to any other clone in the analysis. They were also identical to each other in the qualitative characters but differed in the quantitative characters. F46 was a new clone in the central region and it was maintained in weedy conditions without mulching. F104 was well maintained, mulched and without weeds. C46 and K104 were treated as members of a single clone in the previous analysis and F46 and F104 most probably belong to this clone also.

FB128 was identical to KB53 in qualitative characters, but differences existed in all the quantitative characters. KB53 was previously shown to be the same clone as KB128. FB128 also belongs to this clone.

7.2.3.2 Similar accessions with different names

23 accessions from the farmers' fields had names not encountered among the accessions in the collections. Four of these accessions have already been shown to be identical to accessions in the collection. F167 and F168 were identical to K37 making Enyeru, Enshenyi and Enyambo synonyms. F171 and F172 which were identical to K74/F74 making Enjagata, Konjowet and Endyabawali synonyms.

12 other clones were found to be very similar either to other accessions from the farmers' fields or to other accessions in the reference collections.

Beer accessions

Six beer accessions were very similar to each other (FB176, FB177, FB178, FB179, FB180, FB181) (Table 7.8). FB179 (Baleka from the west), FB180 (Omuzo from the south west) and FB181 (Nakanyala) were small statured like KB130 (Nalusi) and were

identical with KB130 in all the qualitative characters but differed in the quantitative characters. Nalusi, Baleka, Omuzo and Nakanyala are probable synonyms.

FB176 (Ensika from south west), FB177 (Engote from south west) and FB178 (Entundu from the south west) were identical to each other and to KB134 (Engumba from the central region which is also the same clone as KB158) (Table 7.8). Ensika, Engote and Entundu are additional synonyms for the clone designated Engumba in chapter 5. All the six accessions (FB176, FB177, FB178, FB179, FB180, FB181) were also similar to KB133 but KB133 had pendulous cylindrical bunches and thus differed from the six accessions.

Musakala accessions

F174 (Namuunga from the east) was identical to C14 (Namunwe from the central region). The name Namuunga relates to one finger just like the name Namunwe.

The two only differed in the quantitative characters. They represent a single clone (Table 7.8).

Nakabululu accessions

F165 (Mukubakkonde from central region) was very similar to a number of Nakabululu accessions but identical only to K118 (Bifusi) (Table 7.8). Mukubakkonde and Bifusi are probable synonyms. The names relate to a fist and refer to the fruits which are short and plump like a fist.

F175 (Mburiondet from the east) was similar to K118, F47 and C47. However, differed from K118 by not having rounded inflated fruits but was identical in all qualitative characters to F47 and C47 (Nakabululu). Mburiondet and Nakabululu are probable synonyms.

Nfuuka accessions

F162 (Ntika from central region) was very similar to K17 (Kabucuragye) and F160 (Katwalo from the central region) (Table 7.8) but F162 differed in bunch shape and finger arrangement in the bunch. It was considered different from K17 and F160. K17 was, however, identical to F160 in all its qualitative characters and the two were considered to be the same clone. Kabucuragye is the name given to this clone. F162 is a clone not represented in the collections.

Finally F166 (Nabusolo from the east) was similar to F32 and C32 (Namwezi from central region) and K33 (Kulwoni from the east as well)(Table 7.8). The two lack anthocyanin pigments in the stem. F166 was identical to F32 and C32 in the qualitative characters. Differences only existed in the quantitative characters. Nabusolo and Namwezi are probable synonyms and Namwezi is retained as the name of the clone. Kulwoni differed in stature from Nabusolo and was considered a different clone.

7.2.3.3 Accessions with names not encountered in the collections

7 accessions had names not encountered in the collections. F163 (Batule from the central region) and F169 (Enyoya from the west) were both subjectively classified as Musakala accessions. F161 (Nakitembe Omusoga from the east), F170 (Kitika from the west) and F182 (Nakitembut) were subjectively classified as Nakitembe accessions. F164 (Nakibira) from central region and F173 (Rwambarara) from the

Table 7.8: Accessions from farmers' fields which are very similar to other accessions with different names.

Accessions being compared	Correlation coefficient	Quantit./qualit. character differences	Conclusion
Beer			
FB176/FB177	0.939	12/0	probably single clone
FB176/FB178	0.898	13/0	probably single clone
FB176/FB179	0.845	13/1	probably different clones
FB176/FB180	0.854	12/1	probably different clones
FB176/FB181	0.799	13/1	probably different clones
FB177/FB178	0.876	13/0	probably single clone
FB177/FB179	0.870	13/1	probably different clones
FB177/FB180	0.911	12/1	probably different clones
FB177/FB181	0.853	13/1	probably different clones
FB178/FB179	0.903	13/1	probably different clones
FB178/FB180	0.925	13/1	probably different clones
FB178/FB181	0.823	13/0	probably single clone
FB179/FB180	0.958	13/0	probably single clone
FB179/FB181	0.931	13/0	probably single clone
FB180/FB181	0.974	11/0	probably single clone
FB177/KB130	0.794	13/1	probably different clones
FB178/KB130	0.801	13/1	probably different clones
FB179/KB130	0.846	13/0	probably single clone
FB180/KB130	0.930	12/0	probably single clone
FB181/KB130	0.812	13/0	probably single clone
FB176/KB133	0.851	12/2	probably different clones
FB177/KB133	0.820	13/2	probably different clones
FB178/KB133	0.873	13/2	probably different clones
FB179/KB133	0.845	13/2	probably different clones
FB180/KB133	0.876	13/2	probably different clones
FB176/KB134	0.919	13/0	probably single clone
FB177/KB134	0.889	12/0	probably single clone
FB178/KB134	0.920	13/0	probably single clone
FB179/KB134	0.835	13/1	probably different clones
FB180/KB134	0.815	13/1	probably different clones
FB181/KB134	0.759	12/1	probably different clones
FB176/FB134	0.944	12/0	probably single clone
FB177/FB134	0.969	13/0	probably single clone
FB178/FB134	0.894	12/0	probably single clone
FB179/FB134	0.869	13/1	probably different clones
FB180/FB134	0.879	13/1	probably different clones
FB181/FB134	0.824	12/1	probably different clones
Musakala			
F174/C14	0.812	13/0	probably single clone
Nakabululu			
F165/C47	0.909	13/1	probably different clones
F165/K50	0.771	13/2	probably different clones
F165/C52	0.761	13/2	probably different clones
F165/K118	0.812	13/0	probably single clone
F165/K122	0.822	13/2	probably different clones
F175/K118	0.878	13/1	probably different clones
F175/F47	0.785	12/1	probably single clone
F175/C47	0.771	13/0	probably single clone
F47/K118	0.827	13/0	probably different clones
Nfuuka			
F160/K17	0.759	13/0	probably single clone
F162/K17	0.851	13/1	probably different clones
F162/F160	0.893	13/0	probably different clones
F166/F32	0.817	13/0	probably single clone
F166/K33	0.857	13/1	probably different clones

western region were subjectively classified as Nfuuka clones. Although some of these accessions seem to be distant from the other accessions in the clone sets where they belong, they clustered with the clone sets to which they were assigned. They were also classified in the same clone sets by classificatory discriminant analysis (Table 7.4). A brief discussion of each of these accessions is given below.

Musakala

F163 (Batule) was an accession from Semuto site, central region. It is claimed

by the farmers to have been introduced from the east. It was more similar to C4 than to any other clone. It was identical to C3, C4, F4 and K13 in all the qualitative characters but very different in the quantitative characters. The correlation values between F163 and C3, C4, F4 and K13 were slightly below those of the known ramets. The Semuto region was too dry for F163 and it was not thriving. It probably belongs to the same clone as C4 and the other accessions (C3, K13). The name Batule is also related to Mudwale and indicates the heaviness of the bunch. Batule is an additional synonym for the clone designated Mudwale.

F169 was another introduced clone from Tanzania. It was growing at Rugaga (site 4). It had lax bunches and long slender fingers like those of Musakala but arranged almost at right angles to the rachis. It had semipersistent floral parts along the rachis, blunt fruit apices which were not found in Musakala clones and the fruit arrangement in the bunch was different from those of Musakala accessions. It was a new clone in the region and not represented in the collection. The bunch looks like an extended bunch of C29 (Kibuzi). It clustered with C14 and F174, but was not the same clone as these.

Nakitembe

F161 (Nakitembe Omusoga) was a typical Nakitembe clone but very robust. The bunch and fruits were long and large in size. The bunch orientation was pendulous. It was indeed a Nakitembe clone but a giant type not met before.

F170 (Kitika) and F182 (Nakitembut) are accessions reported by farmers to be similar to C27 (Nakyetengu) but growing in the western and eastern highlands respectively. In fact in some areas of the central region Nakyetengu is called Kitika. F170 and F182 are commercial clones in the highlands and they were very well maintained. They were short, like Nakyetengu, with heavy bunches. The two take a long time to drop most of their neuter flowers because of lower temperatures in the highlands. It is common for C27 to have semi-persistent flowers along the rachis. F170 and F182 seemed to survive better at higher altitudes. Farmers in the west mentioned that Kitika (F170) originated from Congo and it is also common in the north western part of Uganda. In the central region the clone is known to have been introduced to the Buganda Kingdom in the 1930s from the west of the country and it was only grown by Nabagereka, the wife of the king of Buganda (farmers' information). It was after some time that ordinary farmers got access to it. It does not thrive well in the central region and therefore C27 looked a bit different from F170 and F182. F182 was most similar to C27 while F170 was most similar to K28. However, K28 was shorter than F182, with persistent floral parts all along its rachis. The three accessions C27, F170 and F182, however, were identical in their qualitative characters and probably represented a single clone. Their differences were due to the growing conditions and different altitudes. Nakyetengu is chosen as the name for this clone.

Nfuuka

F164 (Nakibira) and F173 (Rwambarara) were both Nfuuka clones but gigantic in size. The fruits were also large. Nakibira had rectangular but very compact bunches with medium but large fingers. The name relates to forest because it is grown near forests and thrives better where forests have been. F173 (Rwambarara) originated from Mbarara in western Uganda. It was similar to F164 in fruit size but had almost cylindrical bunches. F164 and F173 were new clones in the collection.

7.2.4 Discussion

The objective of this phase was firstly to find out whether farmers' clones could be placed with accuracy in their appropriate clone sets despite being maintained under different growing conditions. Secondly, it was the aim of this study to assess whether or not all these farmers' clones were represented in the collection.

Different growing conditions in farmers' fields did not prevent farmers' clones being placed accurately in the previously identified clone sets of the Highland bananas. Results however indicated that some differences in between ramets of the same clone could be due to growing conditions in different environments. This point was illustrated by lower average correlation values by higher altitude accessions than those grown in lower altitudes. Differences in higher altitude clones have been observed by farmers and traders for sometime. In Uganda market vendors distinguish similar clones from lower altitude and those from higher altitude zones. Higher altitude clones are known by farmers to be bulky in size of fruits and bunches but much of the content is water not starch. The food therefore is waterly, light yellow and not tasty. The difference therefore noted in this study is that there were large differences in size of all parts which were measured in accessions from higher altitude zones. Size differences in clones grown in higher altitude zones could be due to the fertility of the volcanic soils, the continuous supply of rain and plenty of mulch which retains much of the moisture in the soil.

Classificatory discriminant analysis placed three accessions (F74, F171, F172) in the Nakitembe clone set but with less than 1.0 probability. The three accessions have already been identified as representatives of a single clone. It is therefore not surprising that they were classified in the same clone set by discriminant analysis. F74 had 67% probability of being a Nakitembe accession, F171 and F172 had 83% probability of being Nakitembe accessions. The three accessions have some characters of Nakitembe and some of Musakala. They have semi-persistent floral parts along the rachis, persistent styles and stamens but their male buds are not imbricate as in most Nakitembe accessions. They however have pendulous bunches with long bottlenecked fruits common among Musakala clones. Hence the uncertainty of their classification. We retain them as Nakitembe accessions.

Three other accessions (F151, F170, F182) were subjectively classified as Nakitembe accessions. In cluster analysis they were not associated with any single clone set. Discriminant analysis placed them under Nakabululu clone set. These are accessions with some characters of Nakabululu, some of Nakitembe and some of Nfuuka clone sets. They however have more characters of Nakabululu than the other two clone sets. F170 and F182 have already been considered to represent a single clone with C27 which had been classified as a Nakabululu clone already. The three accessions are retained as Nakabululu accessions.

Table 7.9: Clones representing farmers' accessions in the national collections.

Clone set	Farmers' accessions	Representative accessions in the national collections
Beer		
	FB128	CB53
	FB134, FB176, FB177, FB178	KB134
	FB179, FB180, FB181	KB130
Musakala		
	F2	C2
	F4, F163	C4
	F7, F143	K6
	F107	K107
	F169	none
	F174	C14
Nakabululu		
	F165	C47
	F47, F175	C47
Nakitembe		
	F73	C19
	F74, F171, F172	K74
	F161	none
Nfuuka		
	F30	K30
	F31, F37, F41, F112, F167,F168	K37
	F32, F166	C32
	F46, F104	K104
	F100	K100
	F110	K110
	F115	K115
	F151	K151
	F156	K156
	F160	C17
	F162	none
	F164	none
	F170, F183	C27
	F173	none

Table 7.10: Number of distinct clones recognised

Clone set	Number of distinct clones in the collections	Number of distinct clones so far recognised in Uganda
Beer	14	14
Musakala	8	9
Nakabululu	11	11
Nakitembe	13	14
Nfuuka	33	36
Total	**79**	**84**

Table 7.9 gives a summary of accessions studied from the farmers' fields to give an assessment of those which were represented in the national collections and those which were not represented in the national collections. 5 clones were considered to be new and were not represented in the national collections. These were F169 from Musakala, F161 from Nakitembe, F162, F164 and F173 from Nfuuka. It is therefore important to decide whether these accessions should be added to the reference collection or the core collection. Since morphological diversity was the major criterion for accessions to be retained in the core collection, these clones should be retained in the core collection.

Table 7.10 gives a summary of distinct clones grown in the national collections and those from the farmers' fields so far recognised in Uganda. There are more clones in the field than in the national collections. This may indicate that there was inadequate sampling as such the complete diversity of the crop is not covered by the national collections. Among the priorities to be set is to make plans for further collecting to make sure gaps are closed. The regions close to the borders between Uganda and Tanzania and between Uganda and Zaire could be targeted.

238 accessions were available for the study including farmers' accessions. The total number of Highland banana clones in East Africa has been estimated to be 45-70 (Baker and Simmonds, 1952; Shepherd, 1957). The number of distinct clones found in this study is higher than the estimate of Baker & Simmonds (1952), given the fact that this study covered Uganda only. This study of the Ugandan Highland bananas was probably based on a larger collection than the one which was available to Baker and Simmonds or Shepherd. However, many accessions were phenotypic duplicates.

111 names were placed in synonymy in the study based on the combined data from the two collections, and 16 names from this study joined the synonymy bringing the number to 127. Phenotypic duplicates may need further critical studies using biochemical and molecular data to confirm that they are genotypically identical.

Farmers' information was found to be essential particularly with regard to the movements and changes that take place among the clones and the distribution and meaning of the local names of the clones. There is need to assemble and make available all existing information given by farmers on the crop. This information is supportive when one comes to make final conclusions on similarities and differences between clones. Now that the farmers' clones have been placed accurately in the clone sets which have been identified, this classification is in place as a reference on which to build further studies to understand fully what is going on in this crop.

Chapter 8

GENERAL DISCUSSION

The purpose of the study reported in this thesis was to determine the amount and structure of variation that existed in the national banana germplasm collections of the East African Highland bananas using morphometric methods. It was also the aim of this study to assess the relative merits of these methods in determining differences between the accessions, to determine the characters most responsible for the variation pattern found in the Highland bananas and to work out a provisional classification and identification system for the accessions. This would facilitate the description and identification of the accessions, provide possible synonymy in the crop and ease communication on the different accessions in the collection and particularly for non-specialists working on the crop in various disciplines. Classifying the East African Highland banana accessions would lead to reduction of unnecessary duplication with regard to collection, conservation and research. Studies on pest and disease resistance could proceed by evaluating representative clones resulting from the proposed classification.

For the purpose of this discussion, we shall consider four important aspects: the methods of analysis; the characters; the classification and identification systems proposed; and recommendations for future work on the Highland bananas.

8.1 Methods of analysis

For more than a decade, researchers have recognised some pattern in the data on the Highland bananas (Shepherd, 1957; Sebasigari, 1987; Rossel & Mbwana, 1991). But there was not yet a way of demonstrating this pattern.

The numerical methods reported in this thesis were able to demonstrate a pattern of variation among the Ugandan Highland bananas which is proving to have some validity (Vuylsteke et al., 1996). Similar techniques have been utilised on different crops and the variation within these crops has been found to have some structure (Small *et al.*, 1976; Pickersgill *et al.*, 1979).

At the time this work was carried out there were some uncertainties as to which classificatory procedures would be most suitable. In numerical studies, the choice of method at each step is critically important (Duncan & Baum, 1981). Three types of numerical methods were applied and these were some of the most widely used (James & McCulloch, 1990). The application of these methods has been probably explored sufficiently for some evaluation of their usefulness to be made. However, as Sokal and Sneath (1963) suggested that although most numerical methods have a long history,

they are still developing and many of the techniques are being tested. In this text therefore, we shall comment briefly on their usefulness and disadvantages.

There was no other way in which morphological data on 238 accessions could be analysed simultaneously in a convenient way as the numerical methods have done. The numerical methods have assisted in projecting the pattern of variation found existing among the East African Highland bananas. This is the pattern which we have been trying to detect and display to others and which can have some value to those working on bananas.

Assessment of similarities between accessions was carried out in two different ways; one by correlation coefficient and another by distance coefficient. The two coefficients give slightly different results according to the way they treat qualitative versus quantitative characters and the size of the differences of states within a character. Each coefficient interpreted similar data differently. This further emphasizes the point which Duncan and Baum (1981) stated that at each stage of numerical analysis, there is a decision to be made depending on the type of data available and the objectives of the study. For best results one must make sure that the right coefficient in relation to the type of data is used. Although correlation and distance coefficients are commonly used on mixed data like those in this study, and the two coefficients were used on both data from Kawanda collection and on the combined data of Kawanda and Kabanyolo, correlation coefficient which is less sensitive to size differences was preferred.

On the data of Kawanda collection, cluster analysis placed a large number of accessions in a manageable number of clusters, which was one of the primary purposes of this study. The difficulty with cluster analysis however, is the choice of method to use. Sokal and Sneath (1963) and Sokal (1986) agree that group average clustering gives the best classification results. It has been argued that numerical methods lack any reliable significance test to prove their accuracy (Williams, 1976). The cophenetic correlation coefficient has been used as a means of evaluating the effectiveness of clustering although Duncan and Baum (1981) argue that it is not appropriate for comparing classifications that have differing numbers of taxa at a particular rank. In this study the three methods of clustering which were applied gave generally similar cluster composition, and UPGMA gave the best results as measured by the cophenetic correlation coefficient. Another check of results of cluster analysis is, to compare results of cluster analysis with those of PCA which gives a spatial relationship of individuals under study (Duncan & Baum, 1981; Radford, 1986; van Hintum, 1995).

So principal component analysis gave a summary of information about the accessions by reducing the dimensionality of the original set of characters and maximising the variance of their linear combinations. PCA produced a pattern of variation among the accessions which was similar to that of cluster analysis in having similar members of accessions in groups as clusters found in cluster analysis.

Five clone sets (Beer, Musakala, Nakabululu, Nakitembe and Nfuuka) emerged from cluster analysis and PCA. The Nfuuka clone set was the largest, most heterogeneous and intergraded with other clone sets, especially Musakala. Nakabululu appeared to be the most distinct clone set. A few Nakitembe accessions were intermediate between Nakabululu and Nakitembe. PCA showed that 26 of the 61 characters used were the most useful in separating accessions into clone sets of the East African Highland bananas along the first four components.

Results from the combined data of Kawanda and Kabanyolo collection gave similar results based on cluster analysis. In this study, some accessions were identified as phenotypic

duplicates but many represented distinct clones. PCA results gave similar clone sets as previously recognised in the analysis of Kawanda collection but there was more overlapping between the clone sets making it impossible to define them more precisely.

Classificatory discriminant analysis was then carried out using the clone sets recognised by the previous analyses and my own experience. Five clone sets were finally recognised and retained. Results of discriminant analysis further indicated that Nfuuka and Nakabululu were the least distant clone sets from each other, Nfuuka and Nakitembe were the next least distant from each other. The Beer clone set was the most distant from all other clone sets.

It is now possible to hypothesise about the phenetic relationship of the different clone sets. Nfuuka being the most heterogeneous clone set and intergrading with other clone sets, could possibly have been the original clone set from which the rest diversified through mutations. The other clone sets would then represent old mutants in various stages of diversification, with Beer clone set being the first to diverge from Nfuuka. Potential future clone sets are Ebihuuna, Namwezi and Siira which form distinct clusters within Nfuuka.

This classification now becomes a reference point which can be used as a basis for further predictions while other data may confirm the validity of the clone sets recognised.

Initial results on the fertility of the East African Highland clones were found to relate to the clone sets so far recognised in this work (Vuylsteke *et al.*, 1996). Results on pest resistance indicated that Siira accessions were the clones most susceptible to weevils (Gold *et al.*, 1996). Farmers' information on maturity periods and food texture of the clones are also related to these clone sets. Nakitembe and Nakabululu clones are quick maturing, they produce suckers profusely and they produce soft textured food. Siira and the majority of Nfuuka clones take a long time to mature, they are slow to produce suckers and they produce hard textured food. Namwezi accessions (one of the distinctive cluster of accessions within the Nfuuka group) are quick maturing and produce soft textured food while Musakala clones are intermediate in relation to these characters.

Although numerical methods have summarised information about accessions in a convenient manner, we lost touch with the actual data. Sometimes transformed characters can be worse or better depending on the aims of the study. Effects of environment could not be easily assessed with transformed data (Williams, 1976). Another drawback of numerical taxonomy is that every time a new clone has to be incorporated, the whole numerical procedure has to be repeated.

Otherwise, numerical methods have so far achieved what traditional taxonomy could not do and that is to summarise information about the large number of accessions and characters and to demonstrate a pattern of variation within the accessions on which one can make further assumptions.

8.2 Characters

Characters form the central core of this work and therefore important to determine the way in which they contributed to the classification and identification of the system presented.

This thesis used morphological characters alone to study variation among the Highland banana clones. The majority of characters used were taxonomic not agronomic. Harlan (1975) argued that it will be breeders who will make use of the classifications of crop germplasm and they will be mainly concerned with genetic compatibility while

morphological characters will be secondary. However, Engels (1986) stated very clearly that the importance of a germplasm collection to a breeding programme is strongly dependent on the availability of accurate descriptions of the accessions and on the taxonomic identification of the germplasm. Although the majority of morphological characters are not important to breeders, the taxonomic identifications to which they lead are important (Pickersgill, 1994). Historically, the easily observable or visible characters of gross morphology have been the basis of most initial classifications and this is not likely to change (Baum, 1981). It was therefore essential to start with morphological characters which offer a meaningful and readily available method of recognition and communication.

There has been urgent need to describe germplasm collections on the basis of morphological characters in order to be aware of the variation seen in these plants or groups of plants. It has been argued that morphological characters become increasingly disappointing in different species because they cannot fully make differences between them (Baum, 1981). However, Hawkes (1988) stated that clear morphological differences can be seen in all plants when they are analysed carefully, the only problem was that they become too numerous and too continuous in their expression. Numerical methods offer ways of treating such characters.

Baum (1981) elaborated upon techniques of observation and data acquisition as important topics that any researcher should consider carefully before assessing morphological similarities and differences between clones, cultivars or species. For example, while characters differentiating clones can easily be obtained from farmers who deal with these clones daily, the time and method of scoring, and the number of states of a character need interpretation before one can score them. Even characters which can easily be defined, take time to score. Besides, in vegetatively propagated plants like bananas, their source of variation being only environmental modification and mutation, the presence of environmentally caused variation within clones becomes a problem in an endeavour to distinguish characters which will give reliable results in different environments.

8.2.1 Character contributions

8.2.1.1 Quantitative characters

As it has already been indicated by various workers (Hintum, 1995; Engels, 1983a), the quantitative characters are greatly affected by the environment. In this study, 13 quantitative characters were used and only four were found useful according to PCA results. These were plant height/girth ratio, petiole length/width ratio, fruit length/width ratio and rachis nodes. The fruit/length width ratio was useful in separating the Musakala clone set. Plant height/girth ratio (below 4.4) and petiole length/width ratio (below 2.4) were lower in dwarf accessions in the different clone sets. Rachis nodes being few was characteristic of clones with long rachises almost touching the ground.

8.2.1.2 Qualitative characters

The value of qualitative characters has been demonstrated by various workers (De Langhe, 1961, 1964; Engels, 1983b). De Langhe (1964) discussed at length some of these characters with respect to environment. He gave seven characters which were useful in identifying clones in the plantain subgroup. Of the seven characters he considered to be useful in identifying plantains, 5 of them were also useful in disitinguishing clones of the East African Highland bananas (Table 8.1).

Table 8.1: Characters found useful in recognising groups (#), identifying clones (@) or diagnostic of individual clones (*). ●Those which had loadings above 0.5 on the first 4 components on the combined data of Kawanda and Kabanyolo collection.

Characters	Useful in identifying groups	Useful in identifying clones	Diagnostic of individual clones	Reference
Plant height/girth ratio		@	*	
Undersheath colour ●			*	1996 IPGRI descriptors
Petiole length/width●			*	
Absence of blotches			*	
Colour of blotches			*	Sebasigari, 1987; Rossel, 1990
Sap colour	#	@		1984 IBPGR descriptors
Sap dripping	#	@		
Suckers with tubular leaves			*	Rossel, 1990
Suckers growing at an angle			*	Sebasigari, 1987
Petiole background colour		@		
Petiole anthocyanin		@		
Petiole length/width ratio		@		
Leaf tip twisted	#			
Leaf colour			*	
Peduncle length/girth ratio			*	
Bunch orientation●	#			1984 IBPGR descriptors; De Langhe, 1961
Bunch length/circumference ratio			*	
Bunch shape●	#			1996 IPGRI descriptors
Bunch compactness●	#			1996 IPGRI descriptors
Fruit arrangement			*	
Fruit fusion			*	
Fruit position in a bunch●	#			De Langhe, 1961
Hand arrangement			*	
Fruit skin colour			*	
Absence of ovules			*	
Pulp colour before maturity	#	@		1984 IBPGR descriptors
Pulp colour after maturity●	#			
Pulp with brown sticky excretions	#			Sebasigari, 1987
Pulp taste	#			Sebasigari, 1987; 1996 IBPGR descriptors
Fruit apex●	#	@		De Langhe, 1961
Style on fruit apex●	#	@		
Type of style●		@		1984 IBPGR descriptors
Persistent staminodes●		@		De Langhe, 1961
Fruit cracking●		@		
Fruit length/width ratio●	#			
Fruit shape●	#	@		
Absence of male bud			*	De Langhe, 1961
Rachis position●	#			1996 IPGRI descriptors
Persistent neutral flowers on rachis●	#	@		1996 IPGRI descriptors

Table 8.1 (Cont'd.)

Characters	Useful in identifying groups	Useful in identifying clones	Diagnostic of individual clones	Reference
Presence or absence of male bud anthocyanin●			*	
Male bud shape●	#	@		1984 IBPGR descriptors
Male bud waxiness●		@		1996 IPGRI descriptors
Male bud apex●	#			
Bract imbrication●	#			
Bract not curling			*	1996 IPGRI descriptors
Compound tepal tubular			*	
Free tepal serrated at basal margins			*	
Filament hooked			*	

8.2.1.2.1 Vegetative characters

The majority of the vegetative characters, scored as qualitative characters were useful in identifying particular clones (Table 8.1). A few of these characters were found to vary in different environments.

Particular shades of green such as the bright green colour of pseudostems or the watery green of upper sheaths, petioles and midribs were only reliable if scored at shooting since they faded with maturity.The glossy pseudostems was invariant, among the East African Highland bananas but useful in separating them from other groups.

Variation in colour of foliage was an important diagnostic character for three clones: Bitambi (C43), a clone with anthocyanin pigmentation in the leaves, Namafura (K100), a clone with glossy leaves, and Nasuuna (K101) with variegated leaves. The intensity of anthocyanin varied with season. During the wet season, the red colour was intense but during the dry season,the colour intensity was low.

Both anthocyanin pigments and the black pigments called melanins occur in various parts of the plant. Various shades of red, brown, bronze and black were observed along the pseudostems, upper sheaths, petioles and midribs. The intense black pigmentation in the stem and the purplish black sheaths, petioles and midribs were considered to be mutations by Simmonds (1966). In each group studied, there were clones with names associated with anthocyanin presence. Nakitembe red, Nakubululu red etc. Anthocyanin presence along the upper sheaths, petioles and midribs was one of the common mutation occurring in each clone set. Similarly there were names also like black Nakabululu. These changes related to colour of vegetative parts in particular clones were common in each clone set except Musakala.

Other vegetative characters, which were diagnostic for different clones but did not involve colour of the plant, were the appearance of suckers and their position in relation to parent, sap colour and dripping. The stoloniferous suckers were diagnostic to Mukazialanda (C8, K8) while suckers with tubular leaves were diagnostic to Katalibwambuzi (CB59, KB59). Sap colour and dripping are important diagnostic characters for the Siira accessions. These are accessions with colourless sap which does not drip readily when the plant is wounded or when a leaf is cut.

The degree of suckering was a useful character according to farmers. It was claimed

by farmers that Nakitembe and Nakabululu clones produced suckers profusely but Nfuuka and Musakala produced suckers at a very low rate. This character was not used in this study because the study period was short.

8.2.1.2.2 Female inflorescence structures

The qualitative characters of the female inflorescence were the most useful. Fifteen of the nineteen characters used were useful in grouping the Highland clones. Then majority of female inflorescence characters were useful in identifying clones as well (Table 8.1). There was a wider variation among the female inflorescence characters than among other parts of the plant.

The majority of the female inflorescence characters were stable while others frequently mutated. Blunt tipped fruits were considered common mutations in the *acuminata* accessions by De Langhe (1964). Blunt tipped fruits were not common in the Musakala group. Only one clone so far has been reported to have these blunt apices in Musakala. Musakala probably being a recent group, has not diversified much like other groups.

Fruit position in a bunch was said to be influenced by internode lengths (Simmonds & Stover, 1987), which in turn are influenced by environment. This was not very obvious during these studies. However, a number of accessions particularly in the Musakala clone set were recorded as very similar but differed only in fruit positions and bunch shapes. Fruit orientation was the major character which varied within the clones of Musakala.

Fruit position was also said to be influenced by the orientation of the bunch (De Langhe, 1961). A pendulous bunch commonly has fruits recurving towards the peduncle or rachis. However, two clones had fruits showing no geotropic reaction although they had pendulous bunches. These were Bikowekowe (K147) and Enyoya (F169).

It was alleged by farmers that pulp colour changes were influenced by the type of soil. Others also claimed that soils in higher altitudes could cause changes to bitter fruit pulp. Most beer clones were located in higher altitude zones where volcanic soils predominate. The effect of soil on fruit pulp needs further critical examination. The pulp colour before (3 months after flowering) and after maturity (just before the whole bunch ripens) was a useful character in differentiating clones as long as scored at the right time.

The persistent style and staminodes could be a confusing character in higher altitude zones, where clones which usually have non-persistent styles and stamens take a long time to drop their styles and staminodes because of slow maturity caused by lower temperatures. This means that one could easily score a clone as having non-persistent style and staminodes, when in fact these are non-persistent.

8.2.1.2.3 Male inflorescence structures

Male inflorescence characters were also useful in the grouping of clones. Fifteen qualitative characters were used, and 5 were useful in grouping clones and 7 were useful in identification (Table 8.1). It is unfortunate that male buds are cut off by the farmers but nevertheless, farmers also know that male buds provide some useful characters in differentiating clones. The orientation of the male rachis, the persistent floral parts along the rachis, the male bud apex, shape and the imbrication were very useful characters in grouping clones. Male bud colour was also useful in identification of clones.

Characters of male flowers were not consistent. Neither the serrated basal margins of the free tepal nor the hooked styles were consistently present in clones which were considered to have them.

The absence of a male bud was considered a mutation (De Langhe, 1961). On a few occasions, a stool of the clone which usually has no male bud, would produce plants with male buds. This is not very common but it happens.

Finally 21 characters were useful in grouping clones (Table 8.1) and these had a loading above 0.5 on the first four principal components of the combined data set from Kawanda and Kabanyolo. Sixteen characters were useful in identification of clones within each clone set and such characters were the common mutations occurring in each clone set like petioles with anthocyanin presence. Some of these characters were useful both in grouping clones and also in identifying clones (Table 8.1). 20 characters were diagnostic to individual clones.

8.3 Classification and identification system

Although the East African Highland bananas are known to occur and been adapted to the East African plateau, a region which extends over western Kenya, Uganda, eastern Zaire, Burundi, Rwanda and mainland Tanzania, their diversification in East Africa needs further clarification. What is being described here is the diversification of the crop in Uganda, one of the major growing areas of the crop. The majority of clones known in Uganda are similar to those grown in Tanzania (Tothill, 1940) but some are different (personal observation). It is therefore important that the Highland bananas are studied throughout their range to know exactly the amount of diversification found in the subgroup.

The Highland bananas were named "Lujugira-Mutika" by Shepherd (1957) to differentiate them from other *Musa* AAA clones. Lujugira is a Luganda name used for a Beer clone in the central region of Uganda. Mutika is a cooking clone from Kenya and could be the clone known as Ntika in the language Luganda in Uganda. The name Lujugira-Mutika covers the beer and cooking clones of East Africa. Shepherd (1957) considered Lujugira-Mutika to be a subgroup. This is a category which is not recognised by the International Code of Nomenclature for Cultivated Plants (Trehane *et al.*, 1995). But, as explained earlier, researchers working on various crops have introduced various new informal categories after being dissatisfied with the limited categories provided by the Code. The Highland bananas are also often and informally referred to as *Musa* AAA-EA after De Langhe (1989).

The morphological description of the Highland bananas has been elaborated on by a number of authors (Baker & Simmonds, 1951; Shepherd, 1957; Sebasigari, 1987, 1990; Rossel & Mbwana, 1991; Karamura & Karamura, 1994). The differences between the East African Highland bananas and other *acuminata* clones are as follows. The East African Highland bananas differ from other *acuminata* triploids by having **pseudostems and petiole bases extensively blotched with black, brown or a mixture of both**. The **pseudostems are very glossy in appearance** with purplish under-sheaths and suckers grow vertically although in a few clones they grow at an angle to the main stem. Variation in plant height depends on clone and environment. A number of clones can grow to a height of more than 6 meters under good growing conditions. In this study the height range was between 2.0 and 5.5 meters. The Highland bananas have **dirty green robust leaves whose lamina is much split along the veins compared to most other bananas. They have a dull brownish purple male bud, with flowers having pink anthers.**

Clones are locally distinguished into two classes: beer and cooking bananas. Beer bananas have an astringent and bitter tasting pulp caused by presence of more tannins than in the cooking clones. On peeling and before maturity, the pulp of beer bananas is cream coloured with brown sticky excretions (Sebasigari, 1987). The pulp can be deep cream or orange at maturity. It is important to note that cooking bananas also have a certain degree of astringency before maturity and their flowers and leaves can also be bitter just like the Beer clones. However, the character of sticky excretions is an important distinguishing character.

Farmers distinguish the beer clones further into those which produce strong, medium and weak beer. This is probably related to the amount of tannin in the fruits. The cooking clones are separated into those which mature quickly and produce soft mashed matooke (Nakitembe and Nakabululu) and the slow to mature clones which make hard mashed matooke (Nfuuka and Siira). A few clones of Nfuuka mature quickly (Namwezi, Enzirabushera, Lusumba), while Musakala clones are intermediate.

In this study the Highland bananas have been kept as a subgroup within a group (AAA) as defined by the International Code of Nomenclature for Cultivated Plants. The smallest distinguishable units in the subgroup are clones. The clones have then been grouped according to their overall morphological similarities to form clone sets. Five clone sets are recognised in this study. A clone set is an informal rank below subgroup but above clones. The clone sets are Beer, Musakala, Nakabululu, Nakitembe and Nfuuka. They can be distingushed using the key below.

8.3.1 Key to six clone sets

1 Pulp bitter and astringent, pulp colour before maturity white with brown sticky excretions, pulp colour after maturity cream with brown sticky excretions _____**Beer**

Pulp insipid, pulp colour before maturity white without brown sticky excretions, pulp colour after maturity cream or brownish orange without brown sticky excretions _____**2**

2 Mainflorescence rachis with persistent neuter flowers, imbricated male bud, persistent dry or fresh style and at times with persistent dry stamens on fruit apices_____**Nakitembe**

Male inflorescence rachis nude or with semi-persistent neuter flowers, male bud not imbricated, commonly no persistent style or stamens on fruit apices _____**3**

3 Bunch orientation subhorizontal, fruits short with length/width ratio less than 3.5 and fruits less than 15 cm long, male buds ovate _____**Nakabululu**

Bunch orientation oblique to pendulous, fruits medium to long with length/width ratio above 3.5 and fruits more than 15 cm long), male bud lanceolate, elliptical, cordate or obovate _____4

4 Bunches mainly truncated or cylindrical, very lax with slender bottle- necked fruits _____**Musakala**

Bunches mainly rectangular, compact with inflated or rounded or rectangular fruits with intermediate shaped apices _____**Nfuuka**

The clone set known as Beer was given this name because the fruits are astringent and bitter and they can only be used for production of beer. The beer clones have characteristically much sap so that most local names relate to this character. Nakitembe means “like Ekitembe”, i.e related to Ensete, due to the persistent floral parts all along the rachis. The name is also related to the verb “hurrying” or quick to mature. The name Nakabululu indicates shortness and compactness of bunches and fruits of the clones. However, the name also means getting ripe at once after harvest. Musakala means laxness, indicating the laxness of the bunches in this clone set. Nfuuka means “I am changing.” Nfuuka was a heterogeneous clone set which could possibly be divided into more clone sets but these were considered insufficiently distinct from the remaining clones of Nfuuka. Nfuuka is a dynamic clone set with some unstable clones.

8.3.2 Formal descriptions of clone sets and keys to clones

8.3.2.1 Beer clone set

There is variation in plant height among Beer clones but the range in this study was from 2.5-4.5 meter, with pseudostem and suckers growing vertically. The clones commonly have intense black pigmentation all along the pseudostem, the intensity being more observable at higher altitudes (above 1400 meters). The rate of production of suckers is low, with some clones having suckers with tubular leaves up to one meter. Leaf length varies with clones and their leaf tips do not twist or fold. The beer clones produce plenty of white sticky sap which drips rapidly on wounding the plant. The bunches are pendulous, oblique or subhorizontal. They can be lax, compact or very compact with various types of fruit apices but commonly blunt types (Plate 1). The fruits are of various lengths, commonly inflated (Plate 1), with characteristically bitter, astringent pulps. The pulp is white with sticky brown excretions before maturity (three months from shooting) and cream with brown sticky excretions across the pulp after maturity. The fruits can have persistent styles or staminodes or none. The orientation of the male inflorescence rachis can be subhorizontal, oblique or pendulous with male buds which can be imbricate or not, rounded, lanceolate, elliptical or cordate and with pointed, intermediate shaped or obtuse apices. The colour of the male buds is purplish brown.

The pattern of variation studied in this clone set showed three types of clones. The first are clones with subhorizontal to oblique compact rounded to rectangular bunches of rounded or rectangular blunt and inflated fruits with or without a persistent fresh style (Plate 1). The second are clones with oblique compact rectangular bunches of rectangular non-inflated fruits with intermediate shaped apices. The third are clones with pendulous and less compact cylindrical bunches of rectangular non-inflated fruits of intermediate shaped apices and with very a long pendulous male inflorescence rachis. Fourteen clones are identified in the key.

Key to Beer clones in Uganda

1 Bunch orientation subhorizontal _____2
Bunch orientation oblique to pendulous _____4

2 Fruits 15 cm to 20 cm long, (fruit length/width ratio 3.5 - 4.4), male bud absent _____**Endirira**
Fruits less than 15 cm long (length/width ratio below 3.5), male bud present _____3

3 Male bud ovate, fruits without persistent style and stamens **Entanga**
Male bud cordate, fruits with fresh persistent style and dry stamens **Bwara**

4 Suckers with tubular leaves **Katalibwambuzi**
Suckers with open scale leaves 5

5 Bunches pendulous; bunch shape cylindrical or truncated 6
Bunches oblique, rectangular or rounded 8

6 Male bud imbricate, purplish green, male rachis with persistent floral parts, fruits long (above 20 cm) with a fresh persitent style **Oruhuuna Beer**
Male bud not imbricate, purplish brown, male rachis nude, fruits medium (less than 20 cm) without a persistent style 7

7 Male inflorescence rachis pendulous, long (almost touching the ground), style of male flower almost hooked **Nalukira**
Male inflorescence rachis oblique or subhorizontal, short (not far from the ground), style of male flower straight **Namunyere**

8 Anthocyanin present in upper sheaths, petioles and midribs 9
Anthocyanin absent in upper sheaths, petioles and midribs 10

9 Bunches rounded, fruits with blunt apices **Bagandeseza**
Bunches rectangular, fruits with intermediate shaped apices **Engambani**

10 Fruits with blunt apices 11
Fruits with intermediate shaped apices 12

11 Bunches rounded, fruits with no persistent dry staminodes **Mende**
Bunches rectangular, fruits with persistent dry staminodes **Namadhi**

12 Plants slender, pseudostem height less than 3 m, pseudostem girth less than 55 cm 13
Plants robust, pseudostem height more than 3 m, pseudostem girth more than 55 cm **Engumba**

13 Bunch length less than 26 cm (bunch length/circumference ratio less than 0.4) Nalusi
Bunch length more than 26 cm (bunch length/circumference ratio more than 0.4 **Nametsi**

8.3.2.2 Musakala clone set

Clones of Musakala vary in height from 2.0-4.5 m. A few clones are semidwarf (below 2.0 m) and have a large pseudostem base, short inter-petiole distances, wide petiole bases and short leaves more than 85 cm wide (Mudwale and Mukazialanda). Plants grow vertically but one clone is known to produce suckers growing at an angle. The whole stool therefore has diverging plants. The rate of production of suckers is low with no clones known to have suckers with tubular leaves. Musakala clones have sparse black pigmentation along the pseudostems. The young unfurled leaf or the mature leaves have a tendency to curve or to twist at their tips. Musakala clones produce milky sap which drips on wounding the plant. Bunches are characteristically pendulous, lax (Plate 2), truncated or cylindrical at times rectangular in shape. The fruits are more than 20 cm long (fruit length/width ratio above 4.5), having bottlenecked tips with no persistent style. The fruit pulp is white before maturity, cream at maturity and never has sticky brown excretions. The flavour is insipid. The male inflorescence rachis is pendulous, nude with a non-imbricate brownish purple male bud which is cordate, lanceolate or oblong with an intermediate shaped or pointed tip.

Variation in Musakala clones is not large. Differences in clones are associated with how far the fruits recurve towards the rachis, giving either truncate, rectangular or somewhat cylindrical bunches which could be more or less lax. Nine distinct clones are recognised in the key.

Key to Musakala clones of Uganda

1 Plants less than 2.5 m tall (height/girth ratio below 4.5), bunches truncated, male bud cordate ____ 2
Plants more than 2.5 m tall (height/girth ratio above 4.5), bunches cylindrical, rectangular, male bud lanceolate, oblong or elliptical ____ 3

2 Suckers growing at an angle from the parent ____ **Mukazialanda**
Suckers growing upright ____ **Mudwale**

3 Bunches with fruits showing no geotropic reaction or spreading out in the bunch ____ 4
Bunches with fruits less strongly recurved towards rachis or strongly recurved towards rachis ____ 6

4 Fruits uniseriate, male bud oblong ____ **Namunwe**
Fruits biseriate, male bud lanceolate ____ 5

5 Fruits almost inflated, tips blunt ____ **Enyoya**
Fruits slender, with bottle-necked tips ____ **Muvubo**

6 Peduncle long (length/girth ratio above 2.5) ____ 7
Peduncle short (length/girth ratio below 2.5) ____ **Muturit**

7 Fruits not strongly recurved towards rachis, bunches more rectangular than truncate or cylindrical ____ **Mayovu**
Fruits strongly recurved towards rachis, bunch shapes more cylindrical than rectangular ____ 8

8 Bunch lax, no gaps between hands within the bunch ____ **Musakala**
Bunches less lax, visible gaps between hands within the bunch, first hand markedly distant from the rest ____ **Lumenyamagali**

8.3.2.3 Nakabululu clone set

Nakabululu clones are generally known to be short with plant height ranging from 1.5-3.5 m. Dwarfism as defined by Simmonds (1966) is common among the Nakabululu clones. Some clones are less than 1.5 m, with short petioles and leaves. Leaves are usually more than 75 cm wide. Pseudostems grow vertically with no suckers with tubular leaves. The plants produce suckers very profusely. Nakabululu clones have sparsely distributed black pigmentation although one mutant is known to have intense black pigmentation all along the pseudostem. The clones have commonly subhorizontal, very compact, rounded bunches (Plate 3) with short fruits less than 15 cm long (fruit length/width ratio below 3.5). The fruits have intermediate shaped apices or blunt apices with no geotropic reaction. The fruits usually lack persistent styles on their apices.

The fruit pulp is white to cream with no brown sticky excretions before maturity and the pulp is orange brown and insipid at maturity. The male inflorescence rachis is oblique, and usually nude although two clones have semi-persistent neuter flowers on their male rachis. The male buds are ovate with an obtuse apex and the bracts are not imbricate.

There are four principal variants in this clone set. The typical Nakabululu has short fruits of intermediate shaped apices; a second variant has blunt fruits and semi-persistent neuter flowers along the rachis, a third variant has semi-persistent neuter flowers along the rachis but no ovules in the fruits, a fourth has pendulous bunches with inflated blunt fruits, semi-persistent neuter flowers on the rachis and rectangular bunches. 11 clones have been identified.

Key to Nakabululu clones of Uganda

1 Male inflorescence rachis with semi-persistent neuter flowers. Fruits are almost blunt, and slightly inflated _____2
Male inflorescence rachis nude, fruits with intermediate shaped apices, not inflated _____3

2 Fruits rounded, with no ovules in their pulp _____**Kaitabunyonyi**
Fruits angular, with ovules in their pulp _____6

3 Fruits inflated, and spreading_____**Bifusi**
Fruits rounded not inflated, irregularly arranged in bunch _____4

4 Pseudostem, upper sheaths and base of petioles covered with intense black pigmentation _____**Kazirakwe**
Pseudostem, upper sheaths and base of petioles covered with sparse black pigmentation _____5

5 Anthocyanin absent in upper sheaths, petioles and midribs _____**Nakabululu**
Anthocyanin present in upper sheaths, petioles and midribs _____**Mukite**

6 Fruits with thick persistent style and staminodes _____7
Fruits with no persistent style or staminodes_____8

7 Pseudostem short (less than 2 m), leaves more than 85 cm (leaf length/girth ratio below 2.5), inter-petiole distance below 6 cm _____**Salalugazi**
Pseudostems tall (more than 2 m), leaves less than 85 cm wide, inter- petiole distance more than 15 cm _____**Butobe**

8 Plants tall (more than 3.5 m), fruit apices blunt, persistent floral parts in the middle of the inflorescence rachis **Kibuzi**
Plants short (less than 3.5 m), fruit apices intermediate shaped, persistent floral parts either at the basal end of the rachis or covering the whole rachis 9

9 Pseudostem less than 1.5 m (height/girth ratio below 4.5), neuter flowers persistent on male inflorescence rachis **Ekitetengwa**
Pseudostem more than 1.5 m (height/girth ratio above 4.5), neuter flowers none or semi-persistent on the male rachis 10

10 Plant height/girth ratio below 2.5 **Nakyetengu**
Plant height/girth ratio above 2.5 **Nakyetengu tall**

8.3.2.4 Nakitembe clone set

The Nakitembe clones vary in height from 1.9 to 5.5 m high. They grow vertically with pseudostems covered with sparsely distributed black pigmentation. The clones produce suckers profusely with no tubular leaves. Leaves are long and narrow (less than 75 cm wide). The clones produce milky sap which drips as soon as the plant is wounded. The clones have oblique to pendulous, lax to compact, rectangular (sometimes cylindrical or truncated) bunches. The fruits are 15-20 cm long (length/width ratio between 3.5-4.4). Fruits have apices which are intermediate to bottlenecked in shape, and may or may not be strongly recurved towards rachis or peduncle. The fruits have a persistent fresh or dry style and dry staminodes. The male inflorescence rachis is oblique with semi-persistent or persistent neuter flowers (Plate 4). The male buds are usually strongly imbricate, but a few clones have a non-imbricate male bud. The male bud apices are intermediate shaped to pointed.

Three principle variants occur in the Nakitembe clone set: clones with oblique bunches of medium rectangular fruits, clones with pendulous bunches and slender strongly recurved fruits and clones with pendulous bunches with slender but not strongly recurved fruits, with a large greenish purple male bud. 14 distinct clones are identified in the key.

Key to Nakitembe clones of Uganda

1 Bunches pendulous, fruits long (length/width ratio above 4.5) with persistent fresh style 2
Bunches oblique, fruits medium (length/width ratio between 3.5-4.4), with persistent dry style 3

2 Male buds imbricate, green red in colour, fingers not strongly recurved in the bunch **Oruhuuna Cooking**
Male buds not imbricate, purplish blue in colour, fingers strongly recurved inside the bunch 12

3 Male bud crimson 4
Male bud purplish brown 5

4 Upper sheaths, petioles and midribs watery green, waxless male bud **Nakitembe-Nakawere**
Upper sheaths, petioles and midribs red, waxy male bud. **Mbwazirume**

5 Fruits blunt 6
Fruits with intermediate shaped apices 8

6 Bunches pendulous, cylindrical _____7
Bunches oblique to pendulous, rectangular _____**Bikowekowe**

7 Fruits long (length/width ratio above 3.5), bunch length/circumference ratio more than 0.5 _____**Nakitembe-Omunyoro**
Fruits medium (length/width ratio below 3.5), bunch length/circumference ratio less than 0.5 _____**Lisindaalo**

8 Plant small (pseudostem less than 3 m tall; pseudostem girth less than 55 cm) _____9
Plant robust (pseudostem more than 3 m tall; pseudostem girth more than 55 cm) _____10

9 Upper sheaths, petiole and midribs purplish brown (bronze) _____**Waikova**
Upper sheaths, petiole and midribs green with no purplish brown pigmentation _____13

10 Petioles and midribs flushed with red anthocyanin _____**Nakitembe Red**
Petioles and midribs green _____11

11 Bunches almost truncate, fruits bottle-necked_____**Nakitembe-Omusoga**
Bunches rectangular, fruits with intermediate shaped apices _____**Nakitembe**

12 Bunches small and rounded _____**Nasaala**
Bunches large almost truncate in shape _____**Enjagata**

13 Fruits with persistent fresh style _____**Enyarutere**
Fruits with persistent dry style _____**Nakitembe-Nakamali**

8.3.2.5 Nfuuka clone set

Nfuuka is the most heterogeneous clone set. It has a wide range of variation among characters studied. The height of clones varies from 1.5 to 5.9 m. Several clones are dwarf (Simmonds, 1966). These have pseudostems less than 2.0 m tall and short petioles and leaves. The majority of clones are tall and slender and the rate of production of suckers is very low. The clones produce much milky sticky sap, although few members have colourless sap which does not drip easily when the plant is wounded. The clones in this set have subhorizontal to oblique bunches. A few clones have pendulous bunches. The bunches are compact, rectangular or cylindrical in shape with medium fruits 15-20 cm long (length/width ratio 3.5-4.4). Fruits are blunt or have apices of intermediate shape, not strongly recurved towards the rachis or with no geotropic response. Fruit tips usually have no persistent style or staminodes, but a few clones have a persistent fresh style and sometimes dry staminodes. The male inflorescence rachis is oblique, nude with a lanceolate or elliptical, not imbricate, male bud with an apex which is pointed or of intermediate shape. The male bud is usually purplish brown, but green-yellow in a few clones.

It is very difficult to summarise the pattern of variation found in the Nfuuka clone set because of its heterogeneity. The Namwezi accessions differ by having pseudostems which are bright coloured some without pigmentation at all or or with pigments either black or brown and with a yellow green male bud or purplish brown. They may eventually be considered a distinct clone set. Other clones have upper sheaths, petioles and midribs all red with anthocyanin pigments. Some of these have colourless sap which does not drip on wounding the plant (Siira accessions). The remaining Nfuuka

clones have the same general structural pattern; oblique rectangular bunches, with differences occurring in fruit skin colour, fruit shape and apices and fruit orientation in the bunch. Plate 5 gives the general structure of the Nfuuka bunches. 36 clones are identified in the key.

Key to Nfuuka clones of Uganda

1 Bunches with fruits having no geotropic reaction, fruits blunt with persistent fresh style and dry staminodes or large rough scar of style or no persistent style and staminode _____2
Bunches with fruits strongly or not strongly recurved towards rachis, fruits with intermediate shaped apices and with no persistent style, staminodes or large rough scar of style _____9

2 Bunches rounded and subhorizontal _____3
Bunches rectangular and oblique _____5

3 Fruits fused _____**Nante**
Fruits not fused _____4

4 Male bud cordate, fruits with a thick freshy persistent style and dry staminode _____**Bukumo**
Male bud lanceolate, fruits with a small freshy persistent style _____**Nambi**

5 Fruits with remains of style and staminodes _____6
Fruits with no remains of style and staminodes_____7

6 Fruits less than 15 cm with no large scar around the style _____**Namakhumbu**
Fruits more than 15 cm with a rough large scar around the style __**Enyabakazi**

7 Fruits rounded, short (less than 18 cm) _____8
Fruits gourd shaped, long (above 18 cm)_____**Lusumba**

8 Bunch length small less than 25cm (bunch length/circumference ratio less than 0.5) _____**Mukaddealikisa**
Bunch length more than 25cm (bunch length/circumference ratio above 0.5) _____**Enyabungere**

9 Pseudostems light coloured _____10
Pseudostems dark coloured _____14

10 Male bud yellow green _____**Kasenene**
Male bud purplish brown_____11

11 Pseudostems with no brown, bronze or black pigmentation _____12
Pseudostems with brown, bronze or black pigmentation_____13

12 Plant height/girth ratio below 4.4, petiole length/width ratio below 2.4 _____**Kulwoni**
Plant height/girth ratio above 4.4, petiole length/width ratio above 2.4 _____**Namwezi**

13 Pseudostems with brown pigmentation _____**Enzirabushera**
Pseudostems with black pigmentation _____**Kaasa**

14 Male bud greenish purple_____**Tereza**

Male bud purplish brown ____ 15

15 Fruits variegated ____ 16
Fruits green ____ 17

16 Leaves variegated ____ **Nasuuna**
Leaves green ____ **Kabende**

17 Fruits glossy green, leaves glossy green ____ **Namafura**
Fruits green, leaves green ____ 18

18 Bunches without internodes, fruit in a continuous spiral ____ **Rwezinga**
Bunches with marked internodes, fruits in clearly distinct hands ____ 19

19 Fruits triangular with pointed apices ____ **Enyamanshari**
Fruits rectangular with intermediate shaped apices ____ 20

20 Petioles and midribs red ____ 21
Petioles and midribs green ____ 25

21 Leaves with anthocyanin pigments ____ **Bitambi**
Leaves without anthocyanin pigments ____ 22

22 Bunches pendulous, plant sap colourless and does not drip readily on wounding the plant ____ **Siira**
Bunches oblique, plant sap milky and drips immediately on wounding the plant ____ 23

23 Pseudostems, upper sheaths covered with intense bronze to black pigmentation ____ **Entukura**
Pseudostems, upper sheaths sparsely covered with black and red pigmentation ____ 24

24 Petiole length/girth ratio above 2.5, leaf internode distance more than 14 cm ____ **Nambokho**
Petiole length/girth ratio below 2.5, leaf internode distance less than 14 cm ____ **Kibalawo**

25 Bunches rounded, not compact, male inflorescence rachis subhorizontal ____ **Nakabinyi**
Bunches rectangular, compact, male inflorescence oblique to pendulous ____ 26

26 Bunches pendulous ____ 27
Bunches oblique ____ 32

27 Bunches with a curve ____ **Nakinyika**
Bunches not curved ____ 28

28 Upper sheaths, petioles and midribs green, plant sap milky and drips on wounding the plant ____ 29
Upper sheaths, petioles and midribs not predominantly green, red in marginal area only, sap colourless and does not drip readily on wounding the plant ____ **Atwalira**

29 Bunches cylindrical ____ 30
Bunches rectangular or truncate ____ 35

30 Male rachis pendulous ____ 31
Male rachis oblique ____ **Kabucuragye**

31 Plants short, less than 3.0 m (height girth ratio below 4.6 ____ **Entazinduka**
Plants tall above 3.0 m (height girth ratio above 4.6) ____ **Namunyere Cooking**

32 Fruit pulp creamy orange at maturity ____ **Nakhaki**
Fruit pulp cream at maturity ____ 33

33 Bunches rectangular ____ 34
Bunches more rounded than rectangular ____ **Tuulatwogere**

34 Fruits large and rounded ____ **Nakibira**
Fruits rectangular ____ **Enyeru**

35 Fruits rectangular, slightly recurved towards the rachis ____ **Ntika**
Fruits slender, less recurved towards the rachis ____ **Rwambarara**

8.3.2.7 Recommendations

Classifications and identification systems need constant reviewing and updating to accomodate changes which take place in the crop. Clones which cannot be keyed to one of the six clone sets will need to be discriminated further using posterior probabilities based on discriminant analysis. It is still not known whether this classification and identification system will work throughout the range of the East African Highland bananas but this system is the basis for future studies.

8.3.3 Summary and suggestions for future work

1) The East African Highland bananas are regarded as a subgroup of *Musa* AAA group. This subgroup is divided into clone sets which have been defined as sets of identifiable clones based on overall similarities of morphological characters.

2) Five clone sets have been identified in the East African Highland bananas based on a mixture of personal experience and numerical taxonomy. They are:

-Beer (astringent and with plenty of sap)

-Musakala (very lax bunched)

-Nakabululu (short and compact bunched)

-Nakitembe (persistent floral parts on fruits and rachis, quick to mature)

-Nfuuka (dynamic and unstable, keeps changing). The words in brackets indicate the meaning of the name.

3) 17 characters were useful in distinguishing these clone sets. These are:

-Persistent neuter flowers along the rachis

-Persistent styles on fruit apices

-Persistent staminodes on fruit apices

-Male bud shape

-Male bud apices

-Male bud bract imbrication

-Male inflorescence rachis orientation

-Bunch orientation

-Degree of bunch compactness
-Bunch shape
-Fruit length/width ratio
-Fruit shape
-Fruit apices
-Fruit orientation in the bunch
-Fruit pulp colour after maturity
-Fruit pulp taste
-Fruit pulp with brown sticky excretions

4) Nakabululu and Nfuuka were the least distinct clone sets. Nfuuka and Nakitembe were found to be the next least distinct to each other. Musakala and Beer were very distinct from each other and from the rest of the clone sets.
5) A key to identify the clone sets has been established based on the seventeen characters.
6) The 238 accessions studied represented 84 distinct clones and a key has been constructed for identifying them.
7) A core collection was proposed consisting of 54 clones selected to represent the variation within the 5 clone sets.

8.3.4 Future work

1. A study of the variation that exists in the East African Highland bananas throughout their range is now necessary in order to know the total diversity within the subgroup.
2. More needs to be learned about the way farmers perceive this crop. Information related to the origins, migration and mutations of clones needs to be obtained from farmers and ways found of analysing this information. This information is vital for selection and breeding, utilization, conservation and management of the crop.
3. It is recommended that a core collection be established for use in demonstrations, research and teaching while a computerised information storage and retrieval system is also needed to maintain information about the national and core collections, to provide quick and reliable identification aids including computer based identification.
4. There is need to expand characterisation and evaluation using advanced methods to facilitate use of the core collection to meet the demands for clones with certain yields, resistance and other quality characteristics. Molecular and biochemical characterisation and evolution in the crop should be studied to understand the pattern of domestication and wild ancestors of the East African clones.



Plate 1. Beer clone set: clone Namadhi, showing the inflated fruits with blunt apices and persistent dry styles and staminodes.



Plate 2. Musakala clone set: clone Mudwale, showing the lax truncate bunch of long slender and bottle-necked fruits



Plate 3. Nakabululu clone set: clone Bifusi, showing the subhorizontal compact and short bunch of short rounded fruits.



Plate 4. Nakitembe clone set: clone Mbwazirume (immature), showing the persistent style on fruits, the persistent neuter flowers along the male rachis and the imbricate male bud.



Plate 5. Nfuuka clone set: clone Enyeru, showing the oblique rectangular and compact bunch, with medium fruits not strongly recurved towards the rachis.

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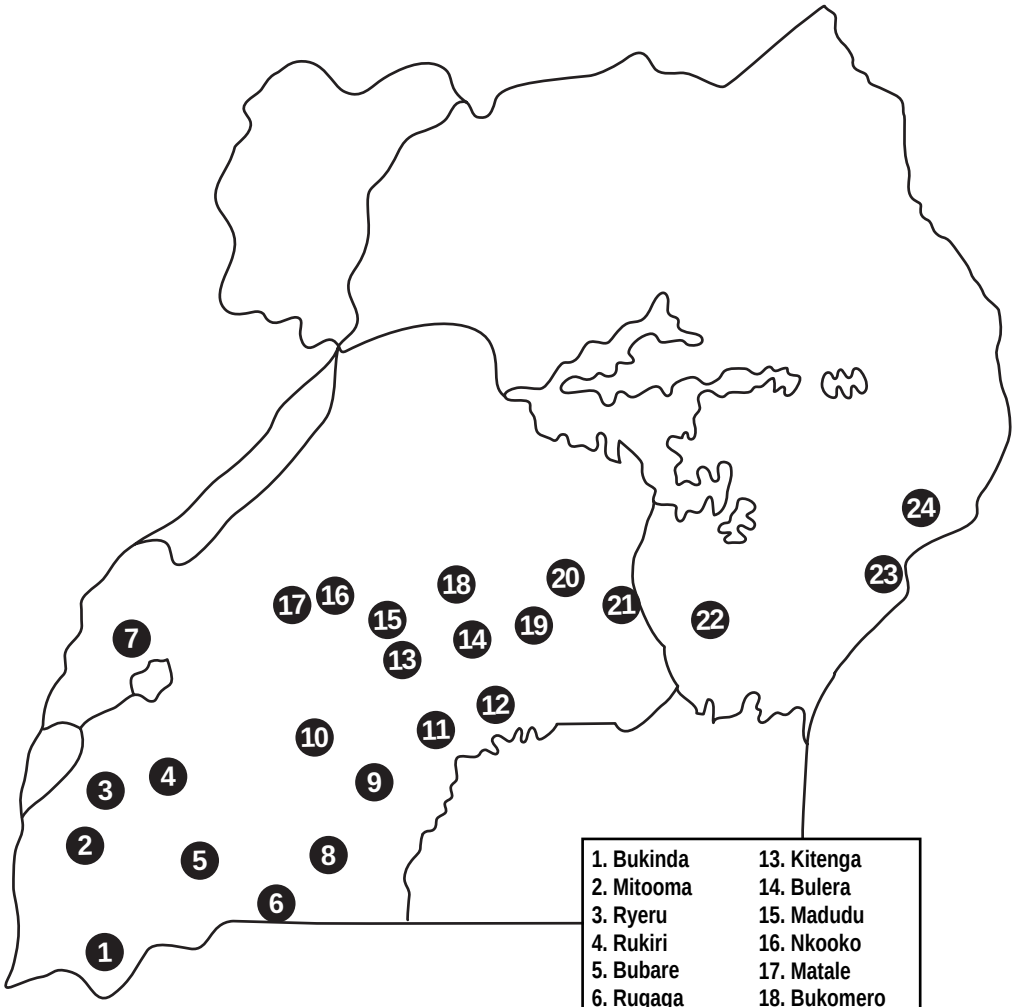
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Appendix1: Standard colours used in character scoring based on the Royal Horticultural Society colour chart

Character	Colour described	Colour standard	Colour chart number
4	Yellow-green	Yellow-green group	144
	Greyed-yellow	Greyed-yellow group	161
7	Brown	Greyed-orange	166
	Bronze	Greyed-purple	187A
	Black	Brown	200
13	Green	Greyed-green	193
	Watery-green	Green group	140
19	Yellow-green	Green group	142
	Dirty-green	Green group	138
	Glossy-green	Cover of green groups	not numbered
33	Waxy-green	Green group	138C
	Green	Green group	139
	Glossy-green	Cover of green group	not numbered
38	White	White group	155B
39	Cream	Orange-White	159
	Orange-brown	Orange group	27A
56	Yellow-green	Yellow-green group	144B
	Crimson	Red-purple group	59
	Purple-blue	Purple group	77
	Bluish-purple	Viola-blue group	not numbered
66	White	White group	155
71	Yellow	Yellow group	4
	Orange	Yellow-orange	23A
	Pink	Red group	56
	Black	Black group	200
73	White	White group	155
	Cream	Yellow group	158A
	Yellow	Yellow group	11B
	Orange	Orange group	27



The location of 24 sites across Uganda at low altitude (sites 11, 16, 18, 20, 21, 22), mid-altitude (3, 5, 8, 9, 10, 12, 13, 14, 15, 17, 19, 23), and high altitude (sites 1, 2, 4, 6, 7, 24) which were used in the diagnostic survey by the Uganda National Programme in 1991 (Gold *et al.*, 1993).

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