Achieving sustainable cultivation of bananas

Volume 2: Germplasm and genetic improvement

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Identifying and classifying banana cultivars

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1 Introduction

The first step in wisdom is to know the things themselves; this notion consists in having a true idea of the objects; objects are distinguished and known by classifying them methodically and giving them appropriate names. Therefore, classification and name-giving will be the foundation of our science.

-Carolus Linnaeus (Linnaeus, 1735).

Taxonomy, otherwise known as systematics, has been described as 'the science of naming, describing and classifying organisms ... using morphological, behavioural, genetic and biochemical observations' (Convention on Biological Diversity, 2019). Taxonomy is the foundation for discussion about the identity and diversity of organisms in the biological world. It provides a means of efficiently retrieving, recording and communicating information about organisms. If a researcher does not have a reliable name for a study object, it is impossible to analyse it accurately and share the results with others. If the taxonomy of a group is poor, this can seriously undermine effective research. If studies are conducted on different taxa (i.e. taxonomic group of any rank such as a species, family or class), but are reported by the same name, this causes confusion and misunderstanding within and between studies.

The practice of classifying and naming organisms enables scientists both to organize large amounts of information about organisms and make deductions about their behaviour based on knowledge of similar organisms. A species' name and position within a larger classification system provides key information about its heritage and helps to explain its characteristics and behaviour. In the case of crops, taxonomy can facilitate the identification, breeding and introduction of new cultivars as well as enforce breeders' intellectual property rights over a new cultivar.

2 Banana cultivars: an overview

Bananas belong to the Musaceae family of flowering plants and the term banana can be used when referring to any members of the family. However, some terms are commonly used to describe additional crop subsets of the family - namely enset, abaca and plantains. Enset and abaca are dealt with a little later. The fruits of plantains are starchy at ripeness and are mostly cooked before consumption. In the narrow sense the term refers to a defined taxonomic grouping (AAB, Plantain subgroup). In the broad sense it contains the latter subgroup as well as several other bananas that are also starchy at ripeness and used for cooking purposes. Where plantain is used in this chapter, it is according to the narrow-sense definition.

Bananas are cultivated around the globe in tropical and subtropical locations and also occur naturally as wild populations of seeded species mainly in Southeast Asia, South Asia and Melanesia. In this chapter our attention is mostly on cultivated bananas. Although the number is somehow variable depending on the source, it is estimated that to date over 1000 banana cultivars occur worldwide, characterized by their own specific morphology (Fig. 1) (Heslop-Harrison and Schwarzacher, 2007). The International Code of Nomenclature for Cultivated Plants (ICNCP) states that cultivars can be characterized as 'assemblages of plants that (a) have been selected for a particular character or combination of characters, (b) are distinct, uniform and stable in those characters, and (c) when propagated by appropriate means, retain those characters' (ICNCP; Brickell et al., 2016). According to the ICNCP, a cultivar name can only be given to cultivated plants whose origin or selection is primarily the result of human interference. People often refer to cultivars as varieties, though this is taxonomically incorrect. Following the ICNCP, a variety is defined as 'a lower rank subdivision of a (wild) species, characterized by a restricted habitat'. However, the usage of the term 'variety' for a cultivar is widespread and unlikely to change. This chapter uses these terms according to their technical ICNCP definitions.



Figure 1 The diversity of fruit characteristics of banana cultivars. (Photo © A. Devouard).

We can easily think of banana cultivars as being just those with edible fruits either cooked or used as a dessert but they can have a broader context. As an example, there are cultivars of enset or Abyssinian bananas (*Ensete ventricosum*) used both as a food and fibre crop, and there are many abaca cultivars (*Musa textilis* and *Musa textilis/balbisiana* hybrids) producing fibre of commercial significance in countries such as the Philippines (Figs. 2 and 3). Some selections of wild species propagated as ornamental bananas (from sections Rhodochlamys and Callimusa in particular) might also be considered as cultivars. EMBRAPA's banana breeding programme in Brazil has an ornamental-breeding component (Fig. 4).

3 Classification at the genera level

Cultivars have been classified (i.e. arranged in categories) based on primary shared characteristics. These categories reflect known or assumed historical relationships. Most of the cultivars (except for Fe'i bananas) are derived from one or two species of the section *Eumusa* (genus *Musa*) of the banana family (Musaceae); *Musa balbisiana* and *Musa acuminata s.l.* (Fig. 5). In understanding taxonomic classification, it is worth remembering Linnaeus' view that 'It is the genus that gives the characters, and not the characters that make the genus' (Linnaeus, 1735). This chapter uses a top-down approach to explore the complexities of banana taxonomy.



Figure 2 Enset, also known as Abysinnian banana (*Ensete ventricosum*), is a staple food for nearly 20% of the Ethiopian population - seen here growing alongside bananas (*Musa* spp.) (E indicates enset, B indicates banana. (Photo courtesy: Guy Blomme, Bioversity International).

All wild and cultivated bananas are herbaceous monocotyledons that belong to the family of Musaceae, a member of the order Zingiberales. Within the Zingiberales, seven additional families are situated: Cannaceae, Heliconiaceae, Costaceae, Marantaceae, Zingiberaceae, Lowiaceae and Strelitziaceae (Kress, 1990). Important features that help to distinguish Musaceae species from the other families are that the stamens of their flowers taken collectively do not



Figure 3 Many cultivars of abaca are of commercial significance in the Philippines. Small bunch of *Musa textilis* showing characteristic male bud with predominant bract imbrication.

have the form or appearance of a petal. In addition, their leaves and bracts are spirally arranged with leaves emerging from the upper part of the pseudostem (Simmonds, 1962).

Musaceae are assumed to be the earliest diversified family within the order Zingiberales (Sass et al., 2016; Janssens et al., 2016; Kress, 1990). They are characterized by many morphological similarities with some of the other early diversified families such as Heliconiaceae and Strelitziaceae. Within the order Zingiberales, a trend can be observed from a large-sized habit and simplified inflorescence (e.g. Heliconiaceae, Musaceae and Strelitziaceae) to a more complex floral organization and a reduced habit for the more recently diversified families in Zingiberales (e.g. Zingiberaceae, Marantaceae, Costaceae and Lowiaceae) (Kirchoff et al., 2009). These differences also help to eliminate confusion with plants from other families in the Zingerberales, notably some *Heliconia* species.



Figure 4 Male buds of hybrids from EMBRAPA's ornamental banana breeding programme demonstrate some of the attractive diversity available of potential cultivars. (Photos courtesy Janay Santos-Serejo).

The Musaceae family contains in total three genera: *Musa*, *Ensete* and *Musella*. However, the taxonomic status of the latter remains to date a matter of dispute, yet is probably congeneric with *Ensete* (Li et al., 2010). From a morphological point of view, several characteristics are available to delineate the different *Musa* genera (Simmonds, 1962). *Musa* are perennial herbs rather



Figure 5 Fe'i bananas are very distinctive – usually with an upright bunch (Karat Kole pictured in Pohnpei, FSM) and pink/purple sap (Toraka Bonubonu pictured in Makira, Solomon Islands).

than monocarpic; their leaf sheaths are tightly clasping instead of rather lax or loosely clasping. Their bracts and flowers are inserted independently on the axis and mostly deciduous by abscission, rather than bracts and flowers being integral with each other and the axis as well as being persistent (deciduous only by rotting, not by abscission).

4 Classification at the section level

Section and/or series (a taxonomic rank lower than section) are typically used to help in the systematic organization of complex genera. Botanists wanting to distinguish groups of species sometimes prefer to create a taxon (a taxonomic group) at the rank of section or series to avoid making new scientific-name combinations which may lead to unnecessary confusion, particularly in cases where taxonomic uncertainties prevail, as is often the case within the genus *Musa*. As one of the leading taxonomists in this field, Simmonds seemed to use the terms sections and series interchangeably over time (e.g. Simmonds, 1954; Stover and Simmonds, 1987).

Within the genus *Musa*, historically four sections have been recognized based on overall morphology, biogeography and cytological data:

- Australimusa;
- Callimusa;
- Eumusa; and
- Rhodochlamys.

More recently, molecular data (Wong et al., 2002; Nwakanma et al., 2003) was used to suggest a reduction from four to two sections in the genus *Musa*. Häkkinen (2013) proposed the two following sections:

- Musa (combining Eumusa and Rhodochlamys); and
- Callimusa (combining former Callimusa, Australimusa and Ingentimusa).

However, more recent molecular phylogenetic research suggests the proposed new arrangement has oversimplified the taxonomic reality. Molecular phylogenetic results (Janssens et al., 2016; Li et al., 2010) have demonstrated that representatives of section Rhodochlamys are scattered among the Eumusa lineages and, as such, should be regarded as polyphyletic. Therefore, from a systematic point of view, it is better to merge the Rhodochlamys representatives in one single natural grouping together with Eumusa species in order to create a more natural classification. Moreover, the polyphyletic nature of Rhodochlamys underlines the huge complexity that is present within the genus as specific morphological characters that were used to discriminate between the two former sections are now proven to contain a high degree of homoplasy. The same molecular phylogenetic studies also showed that the situation of former sections Callimusa and Australimusa is completely different. Results indicate that former section Australimusa can be regarded as a monophyletic clade, positioned among the representatives of former section Callimusa (incl. *M. ingens* as member of Ingentimusa), making the latter a paraphyletic grouping. Following these results, we can assume that the most recent common ancestor of the former section Australimusa is characterized by a unique set of morphological characters, chromosome number and biogeography and that, likewise, a significant degree of homology has been assessed for this group. Moreover, the same is true for members of the former Callimusa section – not taking the position of *M. ingens* into account – with the main difference that Callimusa does not form a monophyletic but a paraphyletic group.

Even though the currently simplified sectional delineation of the genus Musa has decreased the degree of artificial groupings within the genus, it also diminished somehow the discriminative power that was initially present in the original system – especially within the current Callimusa section (Jones and Daniells, 2019). In addition, from a taxonomical perspective, using Musa as a new section name unfortunately promotes unnecessary confusion as now both section and genus share the same name. It is clear that a more detailed study needs to be undertaken before the four-section approach is completely abandoned (Ploetz et al., 2007).

5 Classification at the species level

The polyphyletic nature of groups and lineages within the genus *Musa* does not stop at the sectional level. It is also present at species level. It is generally known that the large diversity in *Musa acuminata* probably led to the immense morphological diversity that is present within cultivated bananas. A number of studies have shown that several of the *M. acuminata* subspecies were involved in the domestication process of bananas (Carreel et al., 2002; Boonruangrod et al., 2009; Perrier et al., 2009).

Based on differences mainly in floral characteristics, multiple subspecies have been recognized in *M. acuminata* of which the following are considered to be taxonomically well-supported: *M. acuminata* ssp. banksii, *M. acuminata* ssp. errans, *M. acuminata* ssp. malaccensis, *M. acuminata* ssp. microcarpa, *M. acuminata* ssp. truncata, *M. acuminata* ssp. zebrina, *M. acuminata* ssp. burmannica and *M. acuminata* ssp. siamea (Simmonds, 1962; Boonruangrod et al., 2008). To date this classification is still a matter of debate as some studies have pointed out that the latter two should probably be merged into one subspecies burmannica (Perrier et al., 2009). Moreover, also *M. acuminata* ssp. microcarpa represents a special case within the *M. acuminata* group as this subspecies is characterized by a large degree of heterozygosity compared

to the other *M. acuminata* subspecies. In fact, its status as subspecies is still unclear as it is often suggested that *M. acuminata* ssp. *microcarpa* populations are possibly the result of hybridization between different gene pools (e.g. subspecies *banksii* and subspecies *zebrina*) and not that the high degree of heterozygosity is caused by the presence of unfixed alleles (Perrier et al., 2009; Martin et al., 2020). Furthermore, based on the results of Janssens et al. (2016) it is clear that subspecies delineation of *Musa acuminata* is rather complicated as there is a clear indication of complex evolutionary patterns and relationships within the species.

The most striking taxonomic discrepancy found within *Musa acuminata* lies in the position of former Rhodochlamys species *M. rosea*, *M. laterita* and *M. rubra*, which fall as a monophyletic group close to *M. burmannica* s.l. (including former subspecies *burmannicoides* and *siamea*). The same situation occurs for the former Rhodochlamys species *M. siamensis* and *M. ornata* that appear to be closely related to *M. acuminata* ssp. *malaccensis*. This indicates that *M. acuminata* s.l. is also a polyphyletic group, which should eventually be taxonomically reassessed after a thorough morphological revision.

Despite its taxonomic complexity, *M. acuminata* s.l. should still be considered as the most important taxonomic unit that has led to the vast number of cultivated bananas currently known to exist. *Musa acuminata* provided the alleged A genome in banana cultivars (discussed below). The species can be discriminated from the other important *Musa* species for banana cultivation (*M. balbisiana*) by having features which include petiole canals with erect or spreading margins, a hairy peduncle and revolute bracts on the male bud (compared to enclosed petiole margins, glabrous peduncle and bracts on the male bud which lift but don't roll). Of the currently recognized subspecies within the *Musa acuminata*, genetic traces of the following four have been found in commercially grown edible banana: *M. acuminata* ssp. *malaccensis, M. acuminata* ssp. *banksii, M. acuminata* ssp. *zebrina* and *M. acuminata* ssp. *burmannica*.

Although the *M. acuminata* subspecies are morphologically rather similar, some clear morphological traits are present to distinguish between them. Especially the position and orientation of the bunch and male rachis as well as bract behaviour can be used to discriminate between the different subspecies, yet even then one has to be well-trained not to make mistakes in the identification of the different subspecies.

6 Current banana cultivar groups

Unaware of the huge diversity of cultivated bananas, Linnaeus applied his binomial system - useful for wild species - to denominate the edible bananas known at that time:

- M. paradisiaca L. (syn. M. cliffortiana; pre-Linnaean name) for plantains; and
- *M. sapientum* L. for dessert and/or cooking bananas.

However, over the years many more cultivars were discovered and it quickly became clear that the use of the original binomial system for the denomination of *Musa* cultivars had become impracticable as different cultivars were assigned under the same name. The discovery of the hybrid origin of cultivated bananas also made clear that the binomial denomination of cultivated bananas was an oversimplification of the complexity present within and among *Musa* cultivars.

The current system of assigning banana cultivars into cultivar groups is based on a system worked out by Simmonds and Shepherd (1955) who understood that the original Latin denomination had become unworkable to further document newly discovered cultivars. Their system was based on two elements

- 1 number of chromosomes (diploid, triploid and tetraploid); and
- 2 the origin of the chromosomes (*M. balbisiana* and/or *M. acuminata* species), where A stands for chromosomes derived from an *M. acuminata* ancestor and B for an *M. balbisiana* ancestor.

However, the distinction into A or B genomes and its associated morphological characterization is now considered as an oversimplification of banana cultivar taxonomy. A full segregation between the different genomes actually does not exist (Baurens et al., 2019), and so it is not fully correct to talk about the different cultivar groups based on chromosome ancestry (e.g. AAB, ABB), which is explained further below. Interestingly in an article by Ken Shepherd (Shepherd, 1990) he says (referring to his paper with Simmonds in 1955) 'The intention then ... was only to elucidate the genomic structure of the cultivars and not to create an international taxonomic tool as the system ... has tended to become.' Nevertheless, despite novel insights in the origin and evolution of banana cultivars in recent years, the system that Simmonds and Shepherd created more than 65 years ago remains in use today to discriminate between Musa cultivars (e.g. Atom et al., 2015) and therefore we will briefly explain their system. When developing their banana cultivar assignment method, Simmonds and Shepherd (1955) selected 15 morphological characters that could be visually assessed and from where a cultivar could be scored into different genome combinations (see Table 1 and Fig. 6).

Their scoring system combined with determination of ploidy gave rise to the different groups of cultivated bananas that we know today:

Character	M. acuminata	M. balbisiana
Pseudostem color	More or less heavily marked with brown or black blotches	Blotches slight or absent
Petiole canal	Margin erect or spreading, with scarious wings below, not clasping pseudostem	Margin enclosed, not winged below, clasping pseudostem
Peduncle Pedicels	Usually downy or hairy Short	Glabrous Long
Ovules	Two regular rows in each loculus (Fig. 6)	Four irregular rows in each loculus (Fig. 6)
Bract shoulder	Usually high (ratio < 0.28 (Fig. 6))	Usually low (ratio > 0.30 (Fig. 6))
Bract curling*	Bracts reflex and roll back after opening (Fig. 6)	Bracts lift but do not roll (Fig. 6)
Bract shape	Lanceolate or narrowly ovate, tapering sharply from the shoulder (Fig. 6)	Broadly ovate, not tapering sharply (Fig. 6)
Bract apex	Acute (Fig. 6)	Obtuse (Fig. 6)
Bract color	Red, dull purple or yellow outside; pink, dull purple or yellow inside	Distinctive brownish- purple outside; bright crimson inside
Color fading	Inside bract color fades to yellow towards the base	Inside bract color continuous to base
Bract scars	Prominent (Fig. 6)	Scarcely prominent (Fig. 6)
Free tepal of male flower	Variably corrugated below tip (Fig. 6)	Rarely corrugated (Fig. 6)
Male flower color Stigma color	Creamy white Orange or rich yellow	Variably flushed with pink Cream, pale yellow or pale pink

Table 1 Characters used in taxonomic scoring of banana cultivars

* In varieties with persistent male bracts, curling is weak or absent, regardless of genotype. *Source*: From Simmonds and Shepherd (1955).

- AA (diploid cultivars, containing genetic material only from M. acuminata);
- AB (diploid cultivars containing genetic material of both *M. acuminata* and *M. balbisiana*);
- AAA (triploid cultivars containing genetic material only from M. acuminata);
- AAB (triploid cultivars, containing genetic material of both *M. acuminata* and *M. balbisiana*, including the Plantain subgroup);
- ABB (triploid cultivars, containing genetic material of both *M. acuminata* and *M. balbisiana*);
- AAAA (tetraploid cultivars containing genetic material only from *M. acuminata*); and
- AAAB, AABB and ABBB (tetraploid cultivars containing genetic material of both *M. acuminata* and *M. balbisiana*).

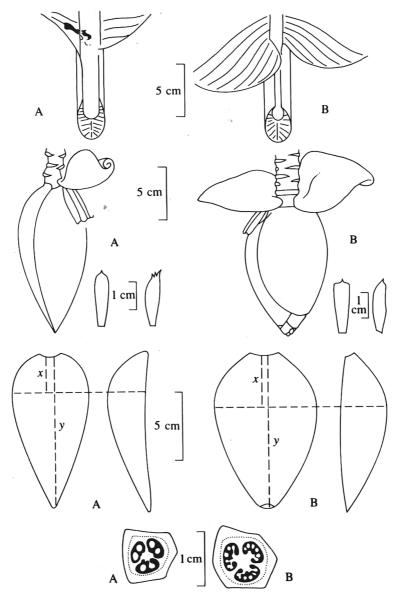


Figure 6 Characteristics of (A) *Musa acuminata* and (B) *Musa balbisiana*: Row 1: petioles; Row 2: male buds (showing shape, bract curling and insertion) and free tepals of male flowers; Row 3: bracts of male buds (ratio = x/y); Row 4: ovule arrangement. (Source: Redrawn from Simmonds and Shepherd, 1955).

If ploidy has been determined by chromosome counts or flow cytometry and the specimen is triploid (either AAA, AAB or ABB) then one can focus on just a few of the descriptors – petiole canal, pseudostem blotching (macules) and internal bract colour to discriminate between the three triploid groups (Shepherd, 1990; Jean-Pierre Horry and Christophe Jenny unpub. pers. comm.).

Within these distinct groups, subgroups are delineated based on morphological characters (e.g. AAA Cavendish subgroup). A subgroup has been identified as: 'A subdivision of a group for a set of cultivars that are assumed to have been generated from a common ancestor by somatic mutation. The ancestor is assumed to be a hybrid between two distinct parents' (MusaNet, 2016). It is important to note that epigenetic variation also plays an important role.

The different cultivars within those subgroups are then given a specific cultivar name (e.g. Williams, Grande Naine, Dwarf Cavendish), which refers to the different clones under which they are traded and grown. So, the formal reference to one of the most cultivated dessert bananas worldwide would then be: *Musa* (AAA Group, Cavendish Subgroup) 'Grande Naine'.

When a new hybrid 'genotype' is created at some point in time (either naturally or via conventional cross-breeding) this hybrid can be denoted as a cultivar when selected for particular characteristics. It can eventually lead to recognition as a subgroup once genetic variants derived from it by mutation or epigenetically are likewise selected as cultivars. Let us suppose, for example, that the first member of what has become the Cavendish subgroup was what is known as Dwarf Cavendish. This has given rise through one or more steps to numerous variants, some of which are named cultivars such as Williams, Formosana and Jaffa. It is important to note that not all the variants would be referred to as cultivars since many (with undesirable characteristics) would be mostly selected against rather than for in breeding or selection programmes.

As alluded to above the time for change has arrived in the way we classify banana cultivars. It is the recent studies on AAB and ABB cultivars using highthroughput sequencing methods which generate whole genome data that showed there is, in fact, no clear segregation between A and B chromosomes (Baurens et al., 2019). Frequent interspecific recombination between both genomes has led to a mosaic genome structure in which elements of the B genome have been introgressed through several generations in the A genome. As such, it is not fully correct to talk about the different cultivar groups based on chromosome ancestry (e.g. AAB, ABB), as a full segregation between the different genomes actually does not exist. A ProMusa-led initiative is currently underway, which in the near future, will set out a suggested way forward in such classifications that both deals with the artificially higher taxonomic ranking conferred upon the current genome groupings and better complies with the requirements of the ICNCP (Vezina, 2019). Meanwhile there is clearly a need for more genetic and genomic research to resolve the ancestry and genetic composition of our banana cultivars.

7 Identifying cultivars

The following section provides practical guidance on identifying cultivars based on the author's experience. How accurate an identification is depends on the quantity and the quality of the plant material and/or information available for the specimen in question and the context in which it is encountered. As with all identification, accuracy depends on experience. Therefore, wherever possible, one should seek advice from experts, either on-site or on-line.

Before one starts the process of identifying a cultivar, one needs to know the exact purpose of the identification required. Is it related to a biosecurity issue, an intellectual property matter or simply to ensure we are talking about the same thing when communicating with others? Other issues include 'How quickly do I need an answer?' 'What resources are available to get the answer?' A shortage of time or money will influence just how much effort can realistically be expended to achieve an answer. As an example, using the 100 or so descriptors in 'Descriptors for Banana' (IPGRI-INIBAP/CIRAD, 1996) requires a massive input of time to get a complete description of a cultivar.

The process to follow also depends on the operator's level of experience. For those with less experience, an important early step is to compare with specimens in a labelled collection. For bananas one ideally needs to grow the specimen alongside reference cultivars. This process takes into account environmental effects on phenotype (the set of observable characteristics of an individual resulting from the interaction of its genotype with the environment). The taxonomic reference collection (http://www.promusa.org/Taxonomic+Re ference+Collection) came about partly to provide such reference cultivars for comparative purposes. Typically herbarium specimens are of limited value for identifying banana cultivars because the vast majority of specimens held are wild species and most herbaria do not give cultivar detail but deal with specieslevel descriptions. Furthermore, because of the immense size of living banana plants and the high moisture content of most plant parts, dried herbarium specimens are relatively of limited use. Often one is required to compare with pictures or descriptions. There are various resources with photo sets available. They include the Musa Germplasm Information System (MGIS https://www .crop-diversity.org/mgis/), which contains key information on Musa germplasm diversity, including passport data, botanical classification, morpho-taxonomic descriptors, molecular studies, plant photographs and GIS information on over 6 500 accessions managed in 29 collections around the world. There are also numerous catalogues available on-line (http://www.musanet.org/).

Identification keys should be consulted where they are available. However, the identification obtained from a key is by no means proof of identity. Keys have their limitations. They are only as good as the information used to create them. They are not necessarily accurate for identification and classification of cultivars

not included in the construction of the key. The other methods discussed can assist in verification. Keys tend to have their best application when developed for regional purposes and where the total number of cultivars is limited.

For those with more experience, a key question is 'Does it look like something I already know?' One can compare what one sees with one's prior experiences. It is about recognizing the whole. This is something like what happens with image recognition technology which uses a range of features to make an identification (e.g. PlantNet https://identify.plantnet.org/). The latter does not cover bananas in detail but is one area worthy of future research/ development for banana identification purposes.

Often the specimen is like something one already knows. One might then have a closer look at a particular characteristic (e.g. male bud shape, bunch and individual fruit characteristics, predominant underlying colour of the pseudostem and presence of particular diseases). Some characteristics are distinctive and can make it easier to distinguish between specimens. The context of the banana is important: 'What is known about the diversity in that location?' It could, for example, be 1 of only 20 cultivars known to occur in that location. Which ones of these are most common? Answering these questions can narrow down the process of identification.

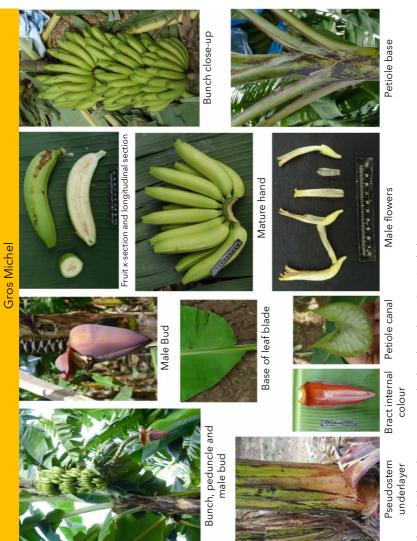
Irrespective of one's level of experience, if the specimen does not appear to be like something one knows, and can't be determined by comparison with pictures, descriptions and keys, then one will need to describe the specimen. Photography is a good starting point for description - 'A picture is worth a thousand words'. If one can only take one photograph, then an in situ photo of a mature bunch with male bud attached would provide the best information for an identification (Fig. 7). If one can take photographs of different features, refer to the 'Guidelines for documenting the minimum set of photos' (https://drive.g oogle.com/file/d/0B6WMCDtu_LjpblZZVktjSWpvdmM/view), which describes how to photograph 14 key characteristics. Figure 8 is an example of photo descriptors applied to the cultivar Gros Michel. The importance of having good photographic skills and equipment cannot be overstated.

Additional information can be gained if one goes beyond descriptive morphology and drills down to analysis at chromosome and molecular level. The ploidy of the specimen can be determined by either flow cytometry, for which a fresh sample of the cigar leaf is required (Fig. 9), or chromosome counting for which root tips are most commonly examined. The cigar leaf can also be used for the molecular characterization. The signature of a specimen at the molecular level then can be compared with an existing database of cultivars such as that of the Musa Genotyping Centre in the Czech Republic (Christelová et al., 2011). Currently microsatellite markers (SSR) can be used to identify to the subgroup level, though identification to the cultivar level is still complex and prohibitively expensive (https://sites.google.com/a/cgxchange.org/musanet/genotyping-centre).



Figure 7 A mature bunch complete with male bud photographed from the side is the single best image to facilitate cultivar identification. Lady Finger (AAB, Pome) pictured in north Queensland.

If time permits more detailed morphological descriptions can be made using the 'Minimum List of Descriptors for Musa' which provides a standardized procedure for morphological characterization of plants based on 34 descriptors with photographs to assist in descriptor scoring (http://www.musalit.org/ seeMore.php?id=14474). Also available are minimum descriptors for East African Highland bananas and plantains (https://sites.google.com/a/cgxchang e.org/musanet/documentation/technical-guidelines). These latter two sets of minimum descriptors were developed in response to the need for additional descriptors, relevant to a particular subgroup to aid in discriminating within the subgroup. MusaTab is an Android© application for recording such Musa characterization data in the field. It allows data to be scored in the field directly on a mobile device and then later uploaded to a computer (https://sites.google .com/a/cgxchange.org/musanet/projects/musatab). Collecting such descriptor



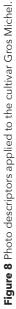




Figure 9 Collecting fresh cigar-leaf (arrowed) samples for both SSR genotyping and determining ploidy can be invaluable for identifying and classifying cultivars (photo courtesy: Agus Sutanto).

information can be a major undertaking and researchers need to appreciate that it is just one of the means to an end and not an end in itself. The end you are mostly seeking is identification and classification of the specimen(s).

MusalD is a taxonomy decision-making application that helps users to identify an unknown taxon by comparison with a reference collection of known Musa taxa described by a set of qualitative descriptors (https://sites.google .com/a/cgxchange.org/musanet/projects/musaid). However, MusalD currently has limitations. These include not having enough accessions in the database and not enough diversity of accessions against which to compare a specimen. There are plans to upgrade the application by adding more characterization data from more diverse genotypes and diverse environments. This should help to validate the methodology behind the tool. The software also needs improvement in its reliability.

In some situations, there may be several cultivars within a subgroup with minor differences that are hard to distinguish. The diversity within the Plantain subgroup is quite immense as well illustrated by the CARBAP collection Musalogue (http://www.musanet.org/). This catalogue contains photos of 145 Plantain accessions which make it possible to identify to the cultivar level in many cases, but in some cases it may only be possible to narrow down the identity of a specimen to a subset of the subgroup, with uncertainty remaining as to the exact cultivar.

8 Future trends and conclusions

There are those who lament the imperfect knowledge existing on banana taxonomy, griping about how confusing it all is to them. However, they fail to realize they can be part of the solution and make a positive contribution by becoming more aware of the information resources nowadays available, utilizing them, and so ensuring in their communication on banana cultivars that they are properly understood by their audiences. The level of global connectedness which the internet has provided can be used to seek assistance/collaboration to solve identification/classification problems together. The recently published CARBAP Plantain collection is a great example of the progress that can be made by a concerted group effort to describe cultivars in detail with good-quality photographs. This has created a platform for better understanding and we look forward to the development of more such cultivar guides. However, like many things it remains a work in progress which can be improved further by linking such information via an on-line key to then more quickly identify an unknown specimen, particularly given the large number (145) of described Plantains it contains.

Recent collecting missions in the Indonesian Triangle (Sutanto et al., 2016) and Bougainville (Sardos et al., 2018) are also good examples of several organizations working together and combining morphological and molecular characterization methods to better understand the wealth of diversity in remote far-off lands while contributing to its conservation for future generations.

Research findings in recent years in banana taxonomy have challenged some of our underlying assumptions regarding cultivar origins and so our knowledge quest is very much ongoing. Additionally, far greater latent variation may be present in certain cultivars than was ever imagined until manifested through agents such as mutagenesis. The broadening of phenotypic diversity by plant improvement programmes, while anticipated to deliver more robust cultivars, may potentially bring additional challenges to our identification efforts.

The 'Global strategy for the conservation and use of *Musa* (banana) genetic resources' (MusaNet, 2016) describes much of the work underway to better understand banana taxonomy and facilitate cultivar identification, as well as additional recommendations for further work. These activities need to continue to be amply resourced with appropriate funding and staffing, including a next-generation well-trained nurtured staff for the task ahead as they are foundational to our broader research efforts on bananas.

9 Where to look for further information

ProMusa is a network of people promoting scientific discussions on bananas. Of particular interest concerning cultivars would be <u>Musapedia</u>: an online, collaboratively built compendium of knowledge on bananas. The ProMusa website is also linked to three databases: <u>Musalit</u>, the largest repository of references on bananas; <u>Musarama</u>, the image bank that provides access to a range of photos on bananas; and <u>Musacontacts</u>, the place to find people working on bananas.

MusaNet is a global network aimed at ensuring the long-term conservation, and facilitating the increased utilization of Musa genetic resources. It provides useful links to Musa collections and diversity publications.

A selection of references illustrating the diversity of cultivars is:

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