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Morphological Taxonomy of *Musa* Genotypes in Taiwan

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This paper presents the morphological classification of 19 *Musa* species and cultivars. Fifteen morphological characters for *Musa acuminata* and *M. balbisiana* and 50 morphological characters adapted from International Union for the Protection of New Varieties of Plant (UPOV) codes were employed to elucidate the phylogenetic relationship between both banana species. Analyses of genetic similarity based on all of these morphological characters suggested that bananas with A–genomes were in the same cluster. Moreover, a genetic similarity coefficient of 0.36 was obtained between *M. acuminata* and *M. balbisiana* in the analysis with the 15 morphological characters for both species, and 0.47 in the analysis with 50 UPOV–based morphological characters. Moreover, principal component analysis (PCA) of the 15 morphological characters suggested that PC1 and PC2 together explained 78.6% of the total variance. A PCA with 50 UPOV–based morphological characters indicated that PC1 explained 80.6% of the total variance, for which the main variables were pseudostem length, leaf blade length, and peduncle length. PCA of the 15 morphological characters showed that 'Pei Chiao', 'Giant Cavendish', and 'Dwarf Cavendish' were proximal in the PCA scatterplot. Notably, the PCA of 50 UPOV–based morphological characters indicated that 'Pei Chiao' and 'Giant Cavendish' were near each other in the PCA scatterplot, suggesting that they are phylogenetically related. The PCA of *M. itinerans* var. *formosana* with 15 morphological descriptors showed that this variant is phylogenetically distant from *M. acuminata* and *M. balbisiana* accessions. In summary, the findings may contribute to the classification and breeding of banana germplasm.

Key words: *Musa*, banana, morphological characterization, Musa, principal component analysis

INTRODUCTION

Banana (*Musa* spp.) is a perennial herbaceous plant of the family Musaceae and a major tropical fruit. Worldwide, the production of banana amounts to 114 million tons, and the area harvested for the fruit is approximately 5 million hectares (FAO, 2014). *Musa* spp. descends from *Musa acuminata* Colla (A genome) and *M. balbisiana* Colla (B genome) and evolve through hybridization into a diploid (AB), triploids (AAA, AAB, and ABB), and tetraploids (AAAA, AAAB, AABB, ABBB) (Simmonds and Shepherd, 1955); commercial banana cultivars are predominantly triploids. Dessert and cooking bananas, both of which are triploids, are one of the world's most important fruit crops, and the Cavendish subgroup, which comprises triploid bananas, accounts for 47% of the global banana production (FAO, 2003). In Taiwan, most banana cultivars are Cavendish banana cv. Formosana, which is locally known as 'Pei Chiao' and belongs to the Cavendish subgroup. Subgroups are cultivated varieties derived from somatic mutation and characterized by little genetic diversity (Channelie're *et al*., 2011). *M. acuminata* includes subspecies and exhibits larger phenotypic variation (De Langhe, 2000).

Bananas are mostly sterile, because they yield fruit by parthenocarpy, produce no seed (Swennen *et al*., 1995). They reproduce asexually, and their somatic mutation occurs separately in different progenitor clones, rendering morphological classification difficult (Onyango *et al*., 2011). Simmonds and Shepherd (1955) established the first morphological characterization of *M. acuminat*a and *M. balbisiana* to classify different bananas in different genome groups, and their characterization system has been widely used (De Langhe, 2000). AA, BB, AB, AAA, AAB, AAAA, AAAB, and AABB bananas, as well as some wild species, are evaluated using quantitative and qualitative descriptors (Ortiz, 1997). IPGRI–INIBAP/CIRAD (1996) has developed descriptors for *Musa* spp. Onyango *et al*. (2011) derived 84 characters of AAB and AA dessert bananas in East Africa from modified descriptors for bananas developed by Bioversity International and CIRAD, and divided *Musa acuminata*–derived cultivars (AAs and AAAs) and hybrids between *M. balbisiana* and *M. acuminata* (AAB) into two separate clusters. They found that 33 of the characters contributed to 71% of the total variance. Descriptors for germplasm characterization are essential to morphological classification, and morphological descriptors should have high heritability and low genotype–by–environment interaction (Ortiz, 1997).

Characterization data can be used in *Musa* conservation; therefore, comprehensively characterizing and documenting *Musa* genetic diversity can contribute to germplasm conservation, the availability of diversity for culti-

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vation, and breeding programs (Channelie're *et al*., 2011). Plants provide essential nutrients for humans. Modern agricultural techniques enable breeders to grow varieties that are resistant to environmental stresses (e.g., pollutants, droughts, and diseases), high–yield, or with superior mouthfeel or enriched color. Breeding renders *Musa* spp. resistant to environmental stress and disease (e.g., *Fusarium* wilt, black leaf streak, and banana streak virus). The genepool provides an abundance of *Musa* germplasm with disease resistances, abiotic stress resistances, and altered agronomic performance (Heslop–Harrison and Schwarzacher, 2007). New cultivars must be patented to safeguard breeders' rights to the cultivars and formulate the rules of usage; therefore, minimum efficient descriptors should be established to facilitate the distinction of the cultivars (Brandao *et al*., 2013). The International Union for the Protection of New Varieties of Plants (UPOV) has established a system of plant variety protection, with the aim of "encouraging the development of new varieties of plants, for the benefit of society" (Byrne, 1993). UPOV (2010) has defined 52 *Musa* spp. characters, and it uses them to identify new banana species and determine their novelty and uniqueness.

The genetic identity and phylogenetic relationship of a cultivar or species are increasingly important to breeders in the terms of protection of their rights to new cultivars or species; moreover, morphological descriptors are preferably applied in varietal identification (Singh *et al*., 2015). The morphological characterization of bananas entails measuring various morphological traits of germplasm collections; univariate analysis and principal component analysis (PCA) are typically performed to identify descriptors that are crucial to characterizing and classifying *Musa* germplasm collections (Ortiz, 1997). Furthermore, PCA is more suitable than univariate analysis for revealing the relationships between characters (Iezzoni and Pritts, 1991). The present study conducted a PCA of the characters of banana germplasm collections in Taiwan to investigate the genetic diversity of the germplasm collections and their phylogenetic relationship.

MATERIALS AND METHODS

Plant materials

This study used 19 *Musa* species and cultivars (two *M. acuminata* materials, two *M. balbisiana* materials, 14 intra– or interspecific hybrids of *M. acuminata* and *M. balbisiana*, and one species of *M. itinerans* var. *formosana*) cultivated in a banana germplasm garden managed by the Taiwan Agricultural Research Institute (Chiayi branch) (Table 1). The geographic coordinates of the garden are 23°48′43″N and 120°46′80″E. It is at an elevation of 33 m with an average annual temperature of 23°C and an average annual precipitation of 2000 mm.

Table 1. Plant materials of genus *Musa* used in this study

No.	Accession ID [a]	Species/hybrid	Genome	Common name	Abbreviation
1	00264380	*M. acuminata*	AA w [b]	*M. acuminata*	ACU
2	00105288	*M. acuminata*	AA cv [b]	Sucrier	SUC
3	00105297	M. acuminata × *M. balbisiana*	AB	Ney Poovan	NPO
4	00105304	*M. balbisiana*	BB w [c]	*M. balbisiana*	BAL
5	00105411	*M. acuminata* (Triploid)	AAA	Dwarf Cavendish	DCA
6	00105466	*M. acuminata* (Triploid)	AAA	Giant Cavendish	GCA
7	00105671	*M. acuminata* (Triploid)	AAA	Morado	MOR
8	00106025	*M. acuminata* (Triploid)	AAA	Pei Chiao	PCH
9	00264577	*M. acuminata* (Triploid)	AAA	Yangambi KM5	YAN
10	00106043	*M. acuminata* × *M. balbisiana* (Triploid)	AAB	Assam	ASS
11	00264700	*M. acuminata* × *M. balbisiana* (Triploid)	AAB	Rilian	RIL
12	00264835	*M. acuminata* × *M. balbisiana* (Triploid)	ABB	Ice Cream	ICR
13	00264844	*M. acuminata* × *M. balbisiana* (Triploid)	ABB	Monkey	MON
14	00264853	*M. balbisiana* (Triploid)	ABB	Nibah	NIB
15	00264335	*M. acuminata* × *M. balbisiana* (Triploid)	ABB	Pelipita	PEL
16	00264826	*M. balbisiana* (Triploid)	BBB	Cooking	COO
17	00264522	*M. acuminata* (Tetraploid)	AAAA	Fhia–17	F–17
18	00264497	*M. acuminata* × *M. balbisiana* (Tetraploid)	AAAB	Fhia–01	F–01
19	00105242	*M. itinerans*	unknown	Formosana	FOR

[a] Accession ID was based on the National Plant Genetic Resources Center of Taiwan Agricultural Research Institute.
[b] *M. acuminata AAw* wild type, *AAcv* cultivar
[c] *M. balbisiana BBw* wild type

Morphological characterization

Simmondsm and Shepherd (1955) proposed 15 morphological characters for *M. acuminata* and *M. balbisiana* (i.e., pseudostem color, petiolar canal, peduncle, pedicel, ovule, bract shoulder, bract curling, bract shape, bract apex, bract color, color fading, bract scar, free tepal of male flower, male flower color, and stigma color) to classify banana cultivars and assess them by the score of 1 (*M. acuminata* character) or 2 (*M. balbiaiana* character). Table 2 and Fig. 1 show the 15 morphological characters.

Fifty morphological characters have been adapted for assessment from UPOV codes for *M. acuminata* Colla; *M.* x *paradisiaca* L. (UPVO, 2010). These characters are presented in Table 3. Among them, 6 are qualitative, 31 quantitative, and 13 pseudo–qualitative. Each of the characters is assessed with a score from 1 to 9.

Data analysis

NTSYSpc 2.2 was used to measure the genetic similarity of samples, the purpose of which was to collect data on their morphological characters (Rohlf, 2006). Next,

Table 2. Characters found most variable based on first four principal components, derived from morphological characters for *M. acuminata* and *M. balbisiana*

Character	Principal components			
	1	2	3	4
Pseudostem color	0.267	0.204	0.309	0.149
Petiolar canal	0.307	–0.096	–0.049	–0.334
Peduncle	0.307	–0.096	–0.049	–0.334
Pedicels	0.348	0.803	–0.402	–0.047
Ovules	0.251	0.086	0.319	–0.327
Bract shoulder	0.022	–0.041	–0.023	–0.015
Bract curling	0.039	–0.055	0.183	–0.014
Bract shape	0.302	–0.129	0.022	0.299
Bract apex	0.265	–0.298	–0.215	0.122
Bract color	0.282	–0.252	–0.150	–0.008
Color fading	0.310	–0.187	–0.133	0.036
Bract scare	0.106	0.121	0.534	0.221
Free tepal of male flower	0.206	0.004	0.447	–0.328
Male flower color	0.282	–0.252	–0.150	–0.008
Stigma color	0.290	0.035	0.097	0.618

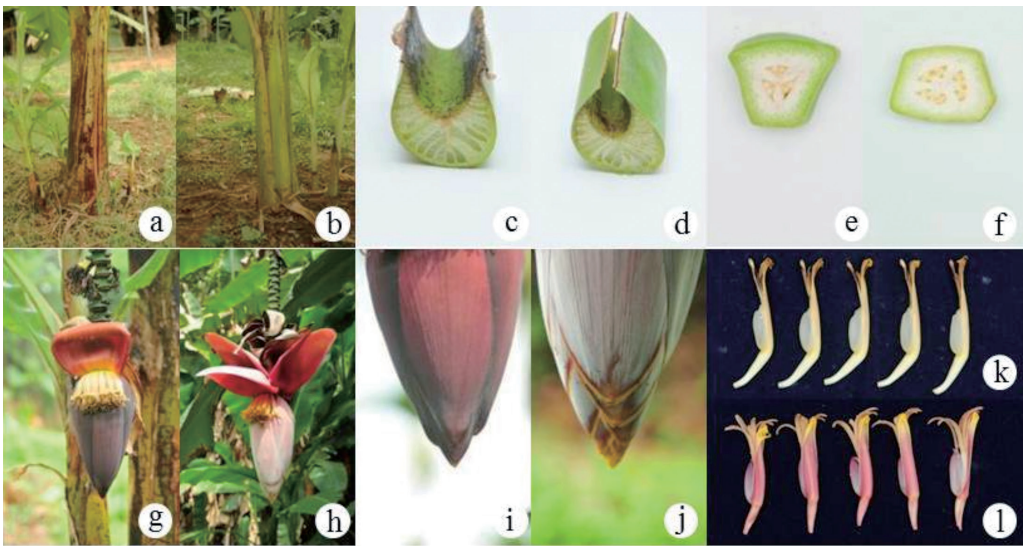

Fig. 1. Morphological traits of various banana accessions used in the scoring of bananas. (a) Pseudostem color: more or less heavily marked with brown of black blotches. (b) Pseudostem color: blotches very slight or absent. (c) Petiole: open canal. (d) Petiole: closed canal. (e) Ovule: two regular rows of ovules. (f) Ovule: four irregular rows of ovules. (g) Bract scare: prominent. Bract: curl. (h) Bract scare: scarcely prominent. Bract: not curl. (i) Bract apex: acute. (j) Bract apex: obtuse. (k) Male flower color: creamy white. (l) Male flower color: variably flushed with pink.

Table 3. Characters found most variable based on first four principal components, derived from morphological characters adapted from the UPOV codes

Character	Principal components			
	1	2	3	4
Ploidy	0.001	0.006	0.007	0.009
Rhizome: number of suckers above ground	0.004	0.016	–0.013	–0.010
Pseudostem: length	0.941	–0.287	–0.134	0.075
Pseudostem: diameter	0.047	0.075	–0.058	0.092
Pseudostem: tapering along length	0	0	0	0
Pseudostem: color	–0.009	–0.012	–0.008	0.014
Pseudostem: intensity of anthocyanin coloration	–0.014	0.025	0.015	–0.022
Pseudostem: color of the inner side of sheath base	0	0	0	0
Plant: compactness of crown	0	0	0	0
Plant: growth habit	0	0	0	0
Petiole: attitude of wings at base	0.009	–0.011	–0.002	0.001
Petiole: length	0.025	–0.099	0.202	–0.267
Leaf blade: color of midrib on lower side	0.001	0	0	0
Leaf blade: shape of base	–0.005	–0.006	–0.004	0.007
Leaf blade: waxiness on lower side	–0.002	0.012	–0.061	0.057
Leaf blade: length	0.282	0.550	0.370	–0.636
Leaf blade: width	0.035	0.374	–0.043	0.124
Leaf blade: ratio length/width	0.002	–0.006	0	0
Leaf blade: glossiness of upper side	–0.007	–0.062	0.057	–0.090
Peduncle: length	0.128	0.348	0.414	0.554
Peduncle: diameter	0.007	0.020	–0.007	–0.004
Peduncle: pubescence	–0.031	0.015	0.014	–0.036
Peduncle: curvature	0.003	0.021	0.013	0.032
Bunch: length	0.099	0.325	0.065	0.338
Bunch: diameter	0.028	0.130	–0.081	0.130
Bunch: shape	0	0	0	0
Bunch: attitude of fruits	0.003	0.006	0.003	0
Bunch: compactness	0	0	0	0
Bunch: number of hands	0.009	0	0	0
Rachis: attitude of male part	–0.003	–0.005	–0.003	0
Rachis: prominence of scars	0.005	0	0	0
Rachis: persistence of bracts	–0.002	0	0	0
Rachis: persistence of hermaphrodite flowers	0.002	0	0.015	0.063
Fruit: longitudinal curvature	–0.003	0.009	0.005	0
Fruit: longitudinal ridges	0.003	–0.007	–0.004	0
Fruit: length	0.007	0.079	–0.033	0.158
Fruit: width (excluding ridges)	0.003	0.008	0.004	0
Fruit: length of pedicel	0.006	0	0	0
Fruit: shape of apex	0	0	0	0
Fruit: thickness of peel	0	0	0	0
Fruit: color of peel (before maturity)	–0.008	0	0	0
Fruit: color of peel	–0.003	–0.034	–0.019	–0.027
Fruit: persistence of floral organs	–0.018	–0.017	0.013	0.001
Fruit: color of flesh	–0.006	0.012	0.006	0
Fruit: firmness of flesh	–0.045	–0.450	0.779	0.106
Male inflorescence: persistence	–0.002	0	0	0
Male inflorescence: shape (in cross section)	0.002	0	0	0
Male inflorescence: overlap of bracts	–0.005	0.007	0	0
Bract: color of inner side	0.002	0.012	0.001	0.041
Male inflorescence: shape of apex of bract	0.008	–0.006	0	0

the unweighted pair–group method with arithmetic mean (UPGMA) was employed to conduct a clustering analysis of their genetic similarity and to plot a phylogenetic dendrogram. PCA was subsequently performed to identify possible relationships between the samples (Iezzoni and Pritts, 1991) and generate biplots in accordance with the morphological characterization of the samples (Osuji *et al*., 1997).

RESULTS

Genetic diversity analysis

Fig. 2a shows the results of the UPGMA analysis of the genetic similarity of the samples based on Simmondsm and Shepherd's (1955) 15 morphological characters of *M. acuminata* and *M. balbisiana*. The results suggested that the eight *M. acuminata* species, three *M. balbisiana* species, and seven of their hybrids belonged to different clusters and had a significant genetic distance (genetic similarity coefficient = 0.36). The analysis yielded two clusters. Cluster 1 comprised 2 subclusters, and Subcluster 1 consisted of *M. acuminata*, 'Sucrier', 'Yangambi KM5', 'Giant Cavendish', 'Dwarf Cavendish', 'Pei Chiao', 'Morado', and 'Fhia–17' (genetic similarity coefficient = 0.85); whereas Subcluster 2 spanned *M. itinerans* var. *formosana* (genetic similarity coefficient = 0.79 with Subcluster 1). Similarly, Cluster 2 comprised 2 subclusters. Subcluster 1 spanned 'Ney Poovan', 'Assam', 'Rilian', and 'Fhia–01' (genetic similarity coefficient = 0.80); whereas Subcluster 2 encompassed *M. balbisiana*, 'Cooking', 'Ice Cream', 'Monkey', 'Nibah', and 'Pelipita' (genetic similarity coefficient = 0.85).

Moreover, all banana species in Subcluster 1 of Cluster 1 belonged to A–genome groups—that is, AA diploids (*M. acuminata* and 'Sucrier'), AAA triploids ('Yangambi KM5', 'Giant Cavendish', 'Dwarf Cavendish', 'Pei Chiao', and 'Morado'), and an AAAA tetraploid (Fhia–17). Except for Moraod, all the other 7 species had a genetic similarity coefficient of 0.93 (Fig. 2a).

Fig. 2b presents the UPGMA analysis results on the genetic similarity of samples based on 50 morphological characters adapted for assessment from the UPOV codes for *M. acuminata* Colla; *M.* x *paradisiaca* L. The results indicated that the respective species of *M. acumi-*

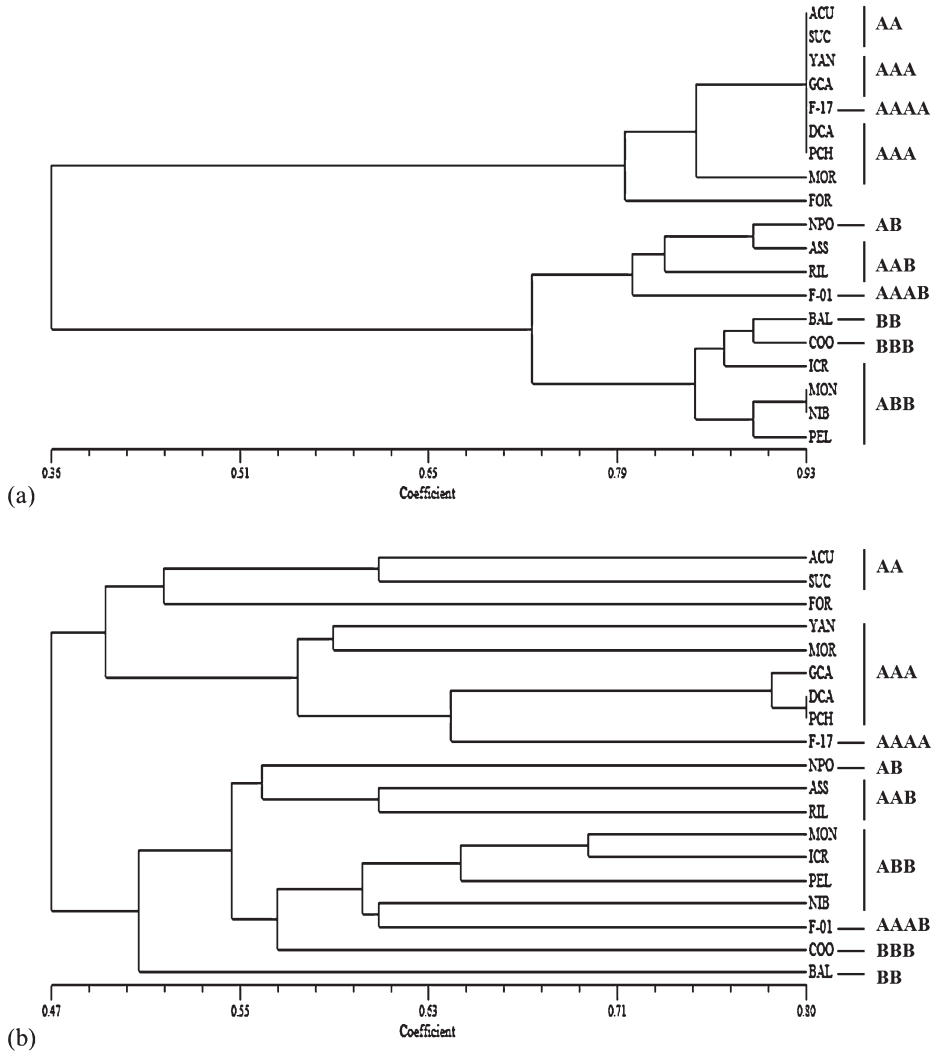

Fig. 2. Dendrogram of genetic similarities in *Musa*, a (above): obtained through morphological characters for *M. acuminata* and *M. balbisiana*, b (below): obtained through morphological characters adapted from the UPOV codes.

nata and *M. balbisiana* and their hybrids belonged to different clusters and had a significant genetic distance (genetic similarity coefficient = 0.47). The analysis yielded two clusters. Cluster 1 comprised 2 subclusters, and Subcluster 1 was divided into two subordinate subclusters. Subordinate subcluster 1 spanned two AA diploids, *M. acuminata* and 'Sucrier' (genetic similarity coefficient = 0.61); whereas Subordinate subcluster 2 included *M. itinerans* var. *formosana* (genetic similarity coefficient = 0.52 with Subordinate subcluster 1). Subcluster 2 consisted of 'Yangambi KM5', 'Morad', 'Giant Cavendish', 'Dwarf Cavendish', 'Pei Chiao', and 'Fhia–17' (genetic similarity coefficient = 0.58). Cluster 2 comprised 3 subclusters. Subcluster 1 spanned 'Ney Poovan', 'Assam', and 'Rilian' (genetic similarity coefficient = 0.56). Subcluster 2 spanned 'Monkey', 'Ice Cream', 'Pelipita', 'Nibah', 'Fhia–01', and 'Cooking' (genetic similarity coefficient = 0.57). Subcluster 3 encompassed a BB diploid, *M. balbisiana* (genetic similarity coefficient = 0.5 with the previous two subclusters).

In addition, all banana species in Subcluster 2 of Cluster 1 belonged to A–genome groups—that is, AAA triploids ('Yangambi KM5', 'Morado', 'Giant Cavendish', 'Dwarf Cavendish', 'Pei Chiao') and an AAAA tetraploid ('Fhia–17') (genetic similarity coefficient = 0.58) (Fig. 2b).

Principal component analysis

The PCA based on the 15 morphological characters of *M. acuminata* and *M. balbisiana* showed that the cumulative variance was 64% for PC1, 78.6% for PC2, 87.6% for PC3, and 91.4% for PC4. PC1 explained 64% of the total variance. Major variables in PC1 were pseudostem color, petiolar canal, peduncle, pedicel, ovule, bract shape, bract apex, bract color, color fading, bract scar, free tepal of male flower, male flower color, and stigma color. PC2 explained 14.6% of the total variance. These results are shown in Table 2. Moreover, the PCA

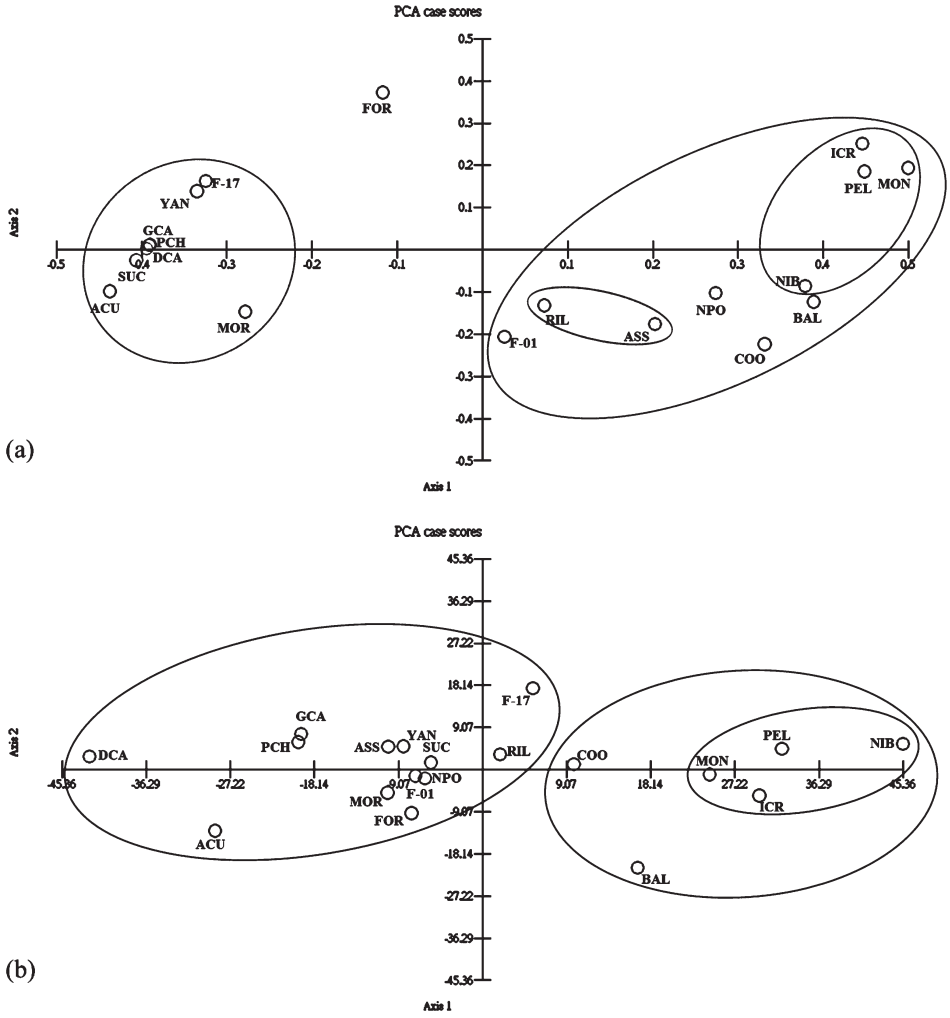

Fig. 3. Scatter plot of the PCA of the 19 *Musa* accessions, a (above): derived from morphological characters for *M. acuminata* and *M. balbisiana*, b (below): derived from morphological characters adapted from the UPOV codes.
(ACU: *M. acuminata*; ASS: 'Assam'; BAL: *M. balbisiana*; COO: 'Cooking'; DCA: 'Dwarf Cavendish'; F–01: 'Fhia–01'; F–17: 'Fhia–17'; FOR: 'Formosana'; GCA: 'Giant Cavendish'; ICR: 'Ice Cream'; MON: 'Monkey'; MOR: 'Morado'; NIB: 'Nibah'; NPO: 'Ney Poovan'; PCH: 'Pei Chiao'; PEL: 'Pelipita'; RIL: 'Rilian'; SUC: 'Sucrier'; YAN: 'Yangambi KM5')

divided the respective species of *M. acuminata* and *M. balbisiana* and their hybrids into two 2–dimensional clusters (Fig. 3a). Cluster 1 spanned AA, AAA, and AAAA accessions (e.g., *M. acuminata*, 'Sucrier', 'Yangambi KM5', 'Morado', 'Pei Chiao', 'Giant Cavendish', 'Dwarf Cavendish', and 'Fhia–17'); in particular, 'Pei Chiao', 'Giant Cavendish', and 'Dwarf Cavendish' were near each other in the PCA scatterplot. Cluster 2 spanned B, BB, AAB, ABB, BBB, and AAAB accessions (e.g., *M. balbisiana*, 'Ney Poovan', 'Assam', 'Rilian', 'Ice Cream', 'Monkey', 'Nibah', 'Pelipita', 'Cooking', and 'Fhia–01'). Notably, 'Assam' and 'Rilian', both of which are AAB triploids, were near each other in the PCA scatterplot. 'Ice Cream', 'Monkey', 'Pelipita', and 'Nibah'—all of which are ABB triploids—were near each other in the PCA scatterplot. *M. itinerans* var. *formosana* did not belong to either cluster.

The PCA based on the 50 morphological characters adapted for assessment from the UPOV codes for *M. acuminata* Colla; *M.* x *paradisiaca* L. indicated that PC1 explained 80.6% of the total variance, accounting for most of the variation in the dataset. Major variables in PC1 were pseudostem length, leaf blade length, and peduncle length. PC2 explained 11.8% of the total variance (cumulative variance = 92.4%). Major variables in PC2 were pseudostem length, leaf blade length, leaf blade width, peduncle length, bunch length, bunch diameter, and fruit flesh firmness. These results are summarized in Table 3. Furthermore, the PCA divided the respective species of *M. acuminata* and *M. balbisiana* and their hybrids into two 2–dimensional clusters (Fig. 3b). Cluster 1 spanned AA, AAA, AAAA, AB, AAB, and AAAB accessions (e.g., *M. acuminata*, 'Sucrier', 'Yangambi KM5', 'Morado', 'Pei Chiao', 'Giant Cavendish', 'Dwarf Cavendish', 'Fhia–17', 'Ney Poovan', 'Assam', 'Rilian', 'Ice Cream', and *M. itinerans* var. *formosana*). Notably, 'Pei Chiao' and 'Giant Cavendish' were near each other in the PCA scatterplot. Cluster 2 spanned BB, ABB, and BBB accessions (e.g., 'Ice Cream', 'Monkey', 'Nibah', 'Pelipita', 'Cooking', and 'Fhia–01'). 'Ice Cream', 'Monkey', 'Nibah', and 'Pelipita'—all of which are ABB triploids—were close to each other in the PCA scatterplot.

DISCUSSION

From the analysis of phylogenetic relationship between 19 *Musa* accessions and the UPGMA analysis of the genetic similarity of the accessions based on the 15 qualitative morphological descriptors for *M. acuminata* and *M. balbisiana*, the results indicate that species in the A–genome groups belong to the same cluster (Fig. 2a). However, AA, AAA, and AAAA accessions (e.g., *M. acuminata*, 'Sucrier', 'Yangambi KM5', 'Pei Chiao', 'Giant Cavendish', 'Dwarf Cavendish', and 'Fhia–17') could not be clustered, and each accession had a genetic similarity coefficient of 0.93. Moreover, the analysis results drawn from the 50 morphological characters adapted from the UPOV codes also suggests that banana species in the A–genome groups were in the same cluster (with AA, AAA, and AAAA accessions divided into distinct subclusters and subordinate subclusters), and AB, BB, AAB, ABB, BBB, and AAAB accession were in another cluster (Fig. 2b). Characterization can be used to assess banana germplasm collections (Nsabimana and Staden, 2005). Therefore, characters of taxonomic relevance that are not subject to environmental factors or the year of cultivation are essential to banana classification (De Langhe *et al*., 2005). Quantitative and multicategory morphoagronomic descriptors are typically adopted to examine the genetic similarity of *Musa* spp. accessions, and reducing the number of descriptors used to characterize *Musa* spp. germplasm does not affect the estimation of genetic variability between *Musa* spp. germplasm (Brandão *et al*., 2013).

The results of the PCA based on the 15 qualitative morphological descriptors showed that PC1 and PC2 cumulatively explained 78.6% of the total variance. Qualitative morphological descriptors have been extensively used to classify different *Musa* germplasm collections under taxonomic groups with similar characters (Simmond and Shepherd, 1955; Tezenas du Montcel *et al*., 1983). Moreover, the results of the PCA with the 50 morphological characters adapted from the UPOV codes indicated that PC1 explained 80.6% of the total variance, and major variables in PC1 were pseudostem length, leaf blade length, and peduncle length. Onyango *et al*. (2011) analyzed 33 characters of AAB and AA dessert bananas in East Africa and found that PC1 and PC2 explained 71% of the total variance; moreover, their findings suggested that morphological characters can be used to distinguish between the genomes and subgroups of *Musa* spp. germplasm and to characterize *Musa* spp. subgroups. Quantitative continuous characters are employed to assess plantains and bananas (Ortiz and Vuylsteke, 1998). Primary quantitative descriptors for *Musa* germplasm collections include pseudostem girth, the number of fruits, and fruit size; such descriptors have high heritability, high repeatability, and low coefficients of variation (Ortiz, 1997).

'Pei Chiao' (Cavendish banana cv. Formosana) (Ko *et al*., 2009), 'Giant Cavendish', and 'Dwarf Cavendish'—all of which are major banana cultivars in Taiwan—are Cavendish varieties with the AAA genome. A UPGMA analysis of the genetic similarity of the three banana cultivars based on the 50 UPOV–derived morphological characters showed that the genetic similarity coefficient was 0.8 for 'Dwarf Cavendish' and 0.78 for 'Giant Cavendish'. Moreover, an analysis of 21 primer pairs of 'Pei Chiao' and 'Dwarf Cavendish' through amplified fragment length polymorphism (AFLP) showed that the genetic similarity coefficient between these cultivars was 0.97. Both share a close phylogenetic relationship with 'Giant Cavendish', the genetic similarity coefficient among three cultivars being 0.99 (Chang *et al*., 2017). Indeed, the PCA based the 15 qualitative morphological characters of suggested that 'Pei Chiao', 'Giant Cavendish', and 'Dwarf Cavendish' were close to each other in the PCA scatterplot (Fig. 3a). However, 'Pei Chiao' and 'Giant Cavendish' were closer to each other in the PCA scatterplot, as the results of a PCA with 50

UPOV–based morphological characters indicated (Fig. 3b). Similar findings were reported by Chang *et al*., (2017). In summary, 'Dwarf Cavendish', 'Pei Chiao', and 'Giant Cavendish' are phylogenetically related.

A genetic similarity analysis based on the 15 qualitative morphological descriptors suggested that *M. itinerans* var. *formosana* and *M. acuminata* are in the same cluster but belong to different subclusters (both subclusters had a genetic similarity coefficient of 0.79); and *M. balbisiana* belonged to a different cluster from *M. itinerans* var. *formosana* and *M. acuminata* (Fig. 2a). In addition, the UPGMA analysis of genetic similarity based on the 50 UPOV–derived morphological characters showed also that *M. itinerans* var. *formosana* was in the same cluster as *M. acuminata* but in a different subordinate subcluster from *M. acuminata* and 'Sucrier'; and both subordinate subclusters had a genetic similarity coefficient of 0.52 (Fig. 2b). These analyses did not group *M. itinerans* var. *formosana* into an independent cluster. However, AFLP can be used to group *M. acuminata*, *M. balbisiana*, and *M. itinerans* into distinct clusters (Chang *et al*., 2017; Wong *et al*., 2002). In summary, this study conducted PCA based on the 15 qualitative descriptors for *M. acuminata* and *M. balbisiana*, the reuslts of which show that *M. itinerans* var. *formosana*, *M. acuminata*, and *M. balbisiana* are in distinct clusters (Fig. 3a), and *M. itinerans* var. *formosana* is phylogenetically distant from the other two banana species.

Morphological, biochemical, and molecular characterization of species or cultivars is widely recognized, and morphological characters are used to quantify the genetic characterization of crops (Singh *et al*., 2015). Multivariate analysis, including PCA and cluster analysis, can be applied in the statistical grouping of germplasm (Ortiz, 1997). The present study found that the 15 morphological characters for *M. acuminata* and *M. balbisiana* and 50 UPOV–based morphological characters can be used to cluster the respective species of *M. acuminata* and *M. balbisiana* and their hybrids. In particular, the PCA based on the 15 morphological characters divided *M. itinerans* var. *formosana* and the respective species of *M. acuminata* and *M. balbisiana* as well as their hybrids into different dimensional clusters. In summary, by comparing genetic diversity between major banana cultivars and wild banana varieties in Taiwan on the basis of morphological characters, the results of this study are expected to inform the classification, breeding, and germplasm conservation of the banana.

AUTHOR CONTRIBUTIONS

1. Shu–Fen CHANG, proved *Musa* species or cultivars of the morphological characters are different, subject to different variables affect the total variance, can analyze the genetic relationship of *Musa* genotypes, and contribute to the classification of banana germplasm.
2. Yung–Fu YEN, designed the genetic diversity analysis for bananas germplasm research under genetic similarity and principal component analysis for studies, and offered suggestion for research.
3. Jer–Way CHANG, offered suggestion on banana germplasm garden managed by the Taiwan Agricultural Research Institute (Chiayi branch).
4. Ikuo MIYAJIMA, offered advices on international tropical horticultural crops research for the morphological classification research, and revised paper and inspected final data.
5. Kuang–Liang HUANG, organized the research protocol through *Musa acuminata* and *M. balbisiana* 15 morphological characters and 50 UPOV–based morphological characters on morphological taxonomy of bananas, and managed lab process.

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