MORPHOMETRIC CHARACTERIZATION AND DIVERSITY ANALYSIS OF A MAKAPUNO COCONUT POPULATION IN U.P. LOS BANOS

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ABSTRACT:- Genetic diversity of embryo cultured makapuno coconut whose endosperm is thicker and softer instead of hard crispy, growing at the University of the Philippines, Los Baños, was assessed using morphometric characters from March 2004 to October 2005. Shannon Weaver Diversity Index and Chi-square (x^2) tests were used to analyze variation in both the qualitative and quantitative characters. Substantial variation in crown shape, bole category, color of young fruit, petiole and inflorescence pigmentation, shape of stem base, fruit characteristics, and leaf structure was observed in the population of 52 palms. Yield of makapuno was estimated as 88%. Simultaneous occurrence of male and female phases allows self-pollination, which favors higher yield of makapuno. The principal component analysis revealed that fruit, nut and husk weight accounted for 99 % of the observed variation. Twelve morphological variables, grouped the population into 6 clusters. Common characters of palm belonging to cluster I are lower weights of fruit, nut and husk with shorter fruit equatorial diameter and shorter internodes. Palms belonging to cluster II have higher weight of nut and longer equatorial diameter. Palms of cluster III are characterized by cylindrical stem and lower number of green leaves. Cluster IV is characterized by heavy fruit and nut weight with elongated shape of nut. Higher number of green leaves presence is the common character of palms of cluster V. Girth at bole region and longer petiole length of leaves are the common characters of palms belonging to cluster VI.

Key Words: Coconut; Makapuno; Genetic Diversity; Quantitative; Qualitative Agronomic Characters; Morphological Characters; Philippines.

INTRODUCTION

Makapuno is a mutant type of coconut, whose endosperm is thicker and softer instead of crispy (Ohler, 1999). It is specifically used for making ice cream, candies, confectioneries, sweetmeats and other food products. Makapuno trait is controlled by a recessive gene (m) and expressed only in homozygous condition (mm), when a makapuno palm (Mm or mm) is pollinated by another makapuno (Mm or mm) and the phenomena is termed as xinia effect. Because of abnormal endosperm, the embryo of makapuno cannot germinate in situ. The makapuno bearing

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trees are seed propagated from the normal nut (Mm) of makapuno bearing palms and as a result, pure makapuno palm (mm) does not exist in nature. The University of the Philippines Makapuno (UPMAC) was the first to develop pure makapuno genotype at the Horticulture Division, University of the Philippines, Los Baños (UPLB) through embryo culture technique (De Guzman, 1970; De Guzman and Del Rosario, 1964; Balaga and De Guzman, 1971). This paved the way to field planting of pure makapuno palms in the Philippines. At UPLB there are 52 embryo cultured makapuno palms, which are reportedly producing as high as 100% makapuno nuts (Cedo et al., 1984). Although each palm in this plantation is homozygous for the makapuno trait, there exists variability with respects to fruit size, color of young fruit, type of makapuno endosperms, inflorescence, floral behavior, and other characters. It is important to know what extent the individual palm differs from one another and to determine the heterogeneity and homogeneity of the population to initiate an effective varietal improvement programme.

MATERIALS AND METHOD

Morphometric Characterization

Each palm of the existing population was characterized morphologically following STANTECH Manual (IPGRI, 1996). A standard color chart was used to document color of flower and fruit. Qualitative stem characters included bole category, shape of stem base, shape of crown, color of leaf-petioles, young inflorescences and young nuts. Bole is the swollen lowermost part of the stem while stem shapes either cylindrical or tapering to about 40 cm above ground. For fruit component analysis, three freshly harvested typical nuts of around 10-11 months old were collected from each tree. Data on types of makapuno and yield (%) were assessed from five consecutive harvests.

There are three types of makapuno

a) Solid endosperm slightly thicker and softer than that of normal coconut; the rest of the cavity is filled with thin liquid endosperm.
b) Soft solid endosperm fills about 50% of the cavity; the rest is filled with thick liquid.
c) The Solid endosperm almost fills the cavity; about a spoonful of thick liquid may be present.

Data of each character was subjected to calculate Chi-square test (x^2c) and Shannon Weaver of Diversity Index (H′) through MS Excel. Principal Component Analysis (PCA) was carried out by SPSS Software. A dendrogram was generated through PCA and the genotypes were grouped into clustering at 1-5 scale of the dendrogram (Figure 1). Diversity indices $H' \geq 0.67$ higher, $H' = 0.34$-0.66 moderate and $H' \leq 0.33$ low (Sourour and Hajer, 2009).

RESULTS AND DISCUSSION

Distribution of Phenotypic Characters

Bole Category and Stem Base

Bole is the swollen lowermost part of the coconut where the roots are generally localized. Three bole categories of palm namely (1) no bole, (2) low bole and (3) high bole were found in the population which distributed equally. The base of coconut is either cylindrical or tapering to about 40cm above ground level. Seventeen percent of the palms in the population have cylindrical stem base while rest had the tapering stem base (Table 1). Bole and stem base distinguish tall from dwarf coconut (Child, 1974). However, the observed distribution of stem base might be due to climatic conditions, cultural practices or both. Heterozygous characters of the parents could have also contributed to the observed variation in the stem base (Foale, 1992).

Crown Shape

Two types of crown namely, spherical and X-shape were observed in the population. Distribution of palms under these two categories of crown was independent and spherical shape of crown was also found dominating over that of X-shape (Table 1). Crown shapes are the result of evolutionary process and adaptation against strong wind. The X-shaped crown could have evolved as a way of minimizing potential wind damage particularly in seashores and elevated hills. Since, the population under study is away from the sea, the observed variation in crown shape might be inherited from the parents of diverse origin. Uddin (2003) reported coconut populations far from the sea to have X-shaped crown too.

Color of Inflorescences and Immature Fruit

Eight distinct inflorescence colors at young stage were identified in the population which were distributed independently. The most abundant inflorescence color in this collection was whitish yellow. Observed colors of immature fruit of the population were green, reddish, reddish green and yellowish green. The distribution of palms for each group of color was statistically different and green color found dominating over other colors (Table 1). Although pigmentation does not improve yield or fruit quality, it can easily be transmitted to the off springs after cross-pollination, thus making it a convenient marker for selection of hybrid at the seedling stage. In addition, bright colors of inflorescence attract insects and birds and thus facilitate cross-pollination and ensure fruit set (Ashburner et al., 2001).

Fruit and Nut Shape

Two kinds of fruit shapes are described of coconut through polar and equatorial views (IPGRI, 1996). There are three descriptive shapes under polar views namely; elliptical, pear and round. Under equatorial view three descriptive shapes are recognized, namely, angular, flat and round. Nut (without husk) shapes may be almost round, round and flat. The distribution of palms producing fruits of elliptical, pear and round shape was uniform while palms having angular, flat and round fruits distributed independently. The distribution of nut shape in the population was also independent (Table 1). In coconut, the fruit or nut is the most economically important part; thus has undergone intensive selection by man. Human selection as well as heterogeneous parent of the population could have contributed to the observed variation in fruit character (Foale, 1992).

Table1. Frequency of coconut palms showing differences in qualitative, vegetative and reproductive characters

Descriptor	Observed Phenotype	Number of Palms Observed	Number of Palms Expected	X^2	Pro-bability	Level of Significance
Bole category (N=52)						
	No bole	22 (42)	17.33			
	Low bole	19 (37)	17.33	3.73	0.15	NS
	High bole	11 (21)	17.33			
Base of the stem (N=52)						
	Tapering	43 (83)	26.00	22.23	0.00	**
	Cylindrical	9 (17)	26.00			
Crown shape (N=52)						
	Spherical	42 (81)	26.00	19.69	0.00	**
	X-shaped	10 (19)	26.00			
Color of young inflorescence (N=42)						
	Cream color	2 (05)	5.25			
	Reddish green	2 (05)	5.25			
	Reddish white	1 (02)	5.25			
	Reddish yellow	1 (02)	5.25	69.24	0.01	*
	Whitish green	2 (05)	5.25			
	Whitish yellow	20 (38)	5.25			
	Yellow	1 (02)	5.25			
	Yellowish green	13 (31)	5.25			
Color of immature fruit (N=50)						
	Green	32 (64)	12.50			
	Reddish	1 (02)	12.50	53.54	0.00	**
	Reddish green	9 (18)	12.50			
	Yellowish green	8 (16)	12.50			
Fruit polar view (N=50)						
	Elliptical	13 (26)	16.67			
	Pear	14 (28)	16.67	3.64	0.30	NS
	Round	23 (23)	16.67			
Fruit equatorial view (N=50)						
	Angular	16 (32)	16.70			
	Flat	4 (08)	16.70	43.28	0.00	**
	Round	30 (60)	16.70			
Nut Shape (N=50)						
	Almost round	27 (54)	16.70			
	Flat	9 (18)	16.70	10.36	0.02	*
	Round	14 (28)	16.70			

*( ) = Percentage of observed value. , * and ** = Significant at 5% and 1% probability, NS = Non significant*

Patterns of Occurrence of Male and Female Phases

Time of occurrence and duration, synchrony of pollen dehiscence (male phase) and female receptivity (female phase) were significantly different among the individual of the population. Overlapping of female phase and male phase was 81% while 19% palms showed no overlapping of male phase and female phases. The highest percent of palms were found

Table 2. Number of palms with open male and receptive female in the same inflorescence and different inflorescence of the same palm

Overlapping of Male and Female Phases	Number of Palms Observed	Expected value	x^2	Probability	Level of Significance
No overlapping	5 (19)	6.75			
Within the inflorescence	2 (70)	6.75	17.59	0.0001	**
Between the inflorescences	16 (59)	6.75			
Within and between the inflorescences	4 (15)	6.75			

*( ) = Percentages of observed values, ** = Significant at 1 % probability*

Table 3. Genotypes producing different types of makapuno

Types of Makapuno	Genotypes	Total	Expected number	Calcu lated x^2	Total x^2	Degree of free-dom (df)	Prob-ability
Producing A, B, C invariably	04, 11, 16, 18, 21, 27, 30, 32, 38, 44, 47, 49, 51, 54, 55, 56	16 (33)	6.63	13.24			
Producing A maximum	02, 05, 09, 13, 26, 33, 34, 43, 53	9 (19)	6.63	0.85			
Producing B maximum	03, 06, 07, 15, 23, 25, 28, 29, 52	9 (19)	6.63	0.85	21.82	6	0.000
Producing C maximum	01, 17, 19, 37, 39, 46, 50	7 (15)	6.63	0.40			
Producing A-B close	14, 36	2 (04)	6.63	0.02			
Producing A-C close	22, 31	2 (04)	6.63	3.23			
Producing B-C close	08, 35, 48,	3 (06)	6.63	3.23			

( ) = Percentages of observed values

Table 4. Shannon Weaver Diversity Index for vegetative and reproductive characters

		Descriptor	SWDI
Vegetative	Qualitative	Bole category	0.52 (M)
		Crown shape	0.60 (M)
		Pigmentation of leaf petiole	0.67 (H)
		Average	**0.60 (M)**
	Quantitative	Girth at bole (20cm above ground level)	0.86 (H)
		Girth at stem (1.5m above ground level)	0.75 (H)
		Length of 11 internodes	0.86 (H)
		Number of green leaves	0.82 (H)
		Leaf Petiole length	0.60 (M)
		Leaf Petiole width	0.56 (M)
		Leaf Petiole thickness	0.62 (M)
		Length of leaf central axis	0.72 (H)
		No. of leaflets	0.61 (M)
		Leaflet length	0.67 (H)
		Leaflet width	0.56 (M)
		Average	**0.69 (H)**
Reproductive	Qualitative	Pigmentation of young inflorescence	0.88 (H)
		Pigmentation of young fruit	0.61 (M)
		Fruit shape (Polar view)	0.66 (M)
		Fruit shape (Equator view)	0.55 (M)
		Nut shape	0.62 (M)
		Average	**0.66 (H)**
	Quantitative	Peduncle length (cm)	0.82 (H)
		Peduncle width (cm)	0.83 (H)
		Peduncle thickness (cm)	0.73 (H)
		Length of central axis (cm)	0.67 (H)
		Spikelet with female flower (FF)	0.71 (H)
		Spikelet without female flower (nos)	0.79 (H)
		Total spikelet (nos)	0.69 (H)
		Total number of button (nos)	0.72 (H)
		Length of spikelet with 1st FF (cm)	0.67 (H)
		Number of bunches	0.69 (H)
		Number of fruits/palm	0.75 (H)
		Average	**0.73 (H)**

M= Medium, H= High

to exhibit male and female phases in between the inflorescences while the least percent of palms were found in occurrence of male phase and stigma receptivity in the same inflorescences (Table 2). Synchrony of pollen dehiscence and female receptivity in a palm enhance occurrence of self-pollination which favored homozygous condition (mm) for makapuno phenotype (Torres, 1937; Zuniga, 1953).

Type of Makapuno

Data recorded from regular harvest during the study period showed that the distribution of palms producing makapuno of all types or combinations of A, B and C types was different from the expected number of palms of equal distribution (Table 3). It was observed that more than 33% of palms invariably producing A, B or C type endosperm. However, palms producing A type endosperm, did not show any B or C types, those producing B type endosperm did not show C or A and those producing C type endosperm had no any harvest of nut possessing A or B type endosperm. Therefore, the population originated from heterozygous and heterogeneous 'Laguna Tall' population. It is expected that individual palms in the present plantation would exhibit high variability in many characters except the makapuno trait. However, the observed variation in makapuno endosperm might be due to the effect of pollen or high heterogeneous nature of the population (Mujer et al., 1984).

Phenotypic Diversity Indices

The estimated diversity indices (SWDI), H´, of vegetative and reproductive morphology of the germplasm indicated relative abundance of their attributes (Table 4 and 5). H´ values of qualitative vegetative traits varied from 0.52 for bole category to 0.67 for pigmentations of leaf petiole with an average of 0.60 indicating the presence of moderate diversity of the attributes. In quantitative vegetative attributes, best indices were estimated for girth of bole at 20 cm above ground level; girth at stem at 1.5 m above ground level; length of 11 leaf scars; number of green leaves and number of fruit per palm. In reproductive qualitative traits, the best indices were estimated in pigmentation of young inflorescence and the lowest in equatorial view of fruit

Table 5. Shannon Weaver Diversity Index of quantitative characters of fruit and nut

Character	SWDI
Fruit	
Weight (g)	0.82 (H)
Polar diameter (cm)	0.79 (H)
Equatorial diameter (cm)	0.75 (H)
Polar circumference (cm)	0.85 (H)
Equator circumference (cm)	0.79 (H)
Average	0.80 (H)
Nut	
Weight (g)	0.81 (H)
Polar diameter (cm)	0.55 (M)
Equatorial diameter (cm)	0.62 (M)
Polar circumference (cm)	0.68 (H)
Equator circumference (cm)	0.75 (H)
Average	0.68 (H)
Fruit Component	
Husk wt (g)	0.87 (H)
Nut/fruit (%)	0.82 (H)
Husk/fruit (%)	0.80 (H)
Average	0.79 (H)

shape with an average of 0.66. High phenotypic diversity indices in reproductive parameters of inflorescences showed an average of 0.73. The best estimate indices were in the length and width of peduncle as well as in the spikelet without female flower (Table 4). Fruit and fruit component analysis showed that the weakest indices 0.55 was for polar diameter and 0.62 for equatorial diameter of nut (Table 5).

Table 6. Eigen values of the Covariance Matrix of 17 principal components of embryo cultured makapuno coconuts at the UPLB

	Eigenvalues	Difference	Proportion	Cumulative
PRIN1	144284	113522	0.820576	0.82058
PRIN2	30762	30398	0.174956	0.99553
PRIN3	364	191	0.002070	0.99858
PRIN4	173	78	0.000983	0.99912
PRIN5	94	26	0.000537	0.99951
PRIN6	69	30	0.000392	0.99973
PRIN7	39	11	0.000220	0.99988
PRIN8	27	15	0.000155	0.99995
PRIN9	13	8	0.000072	0.99998
PRIN10	5	3	0.000026	0.99999
PRIN11	2	1	0.000010	1.00000
PRIN12	1	1	0.000006	1.00000
PRIN13	0	0	0.000002	1.00000

Table 7. Intra and inter cluster distances among the various clusters of embryo cultured makapuno coconuts

Cluster	I	II	III	IV	V	VI
I	163.58	1194.041	485.729	1603.219	821.52	882.992
II		110.19	748.250	507.076	375.196	474.420
III			128.58	1122.836	397.946	400.567
IV				139.37	830.952	737.907
V					146.30	326.024
VI						146.21

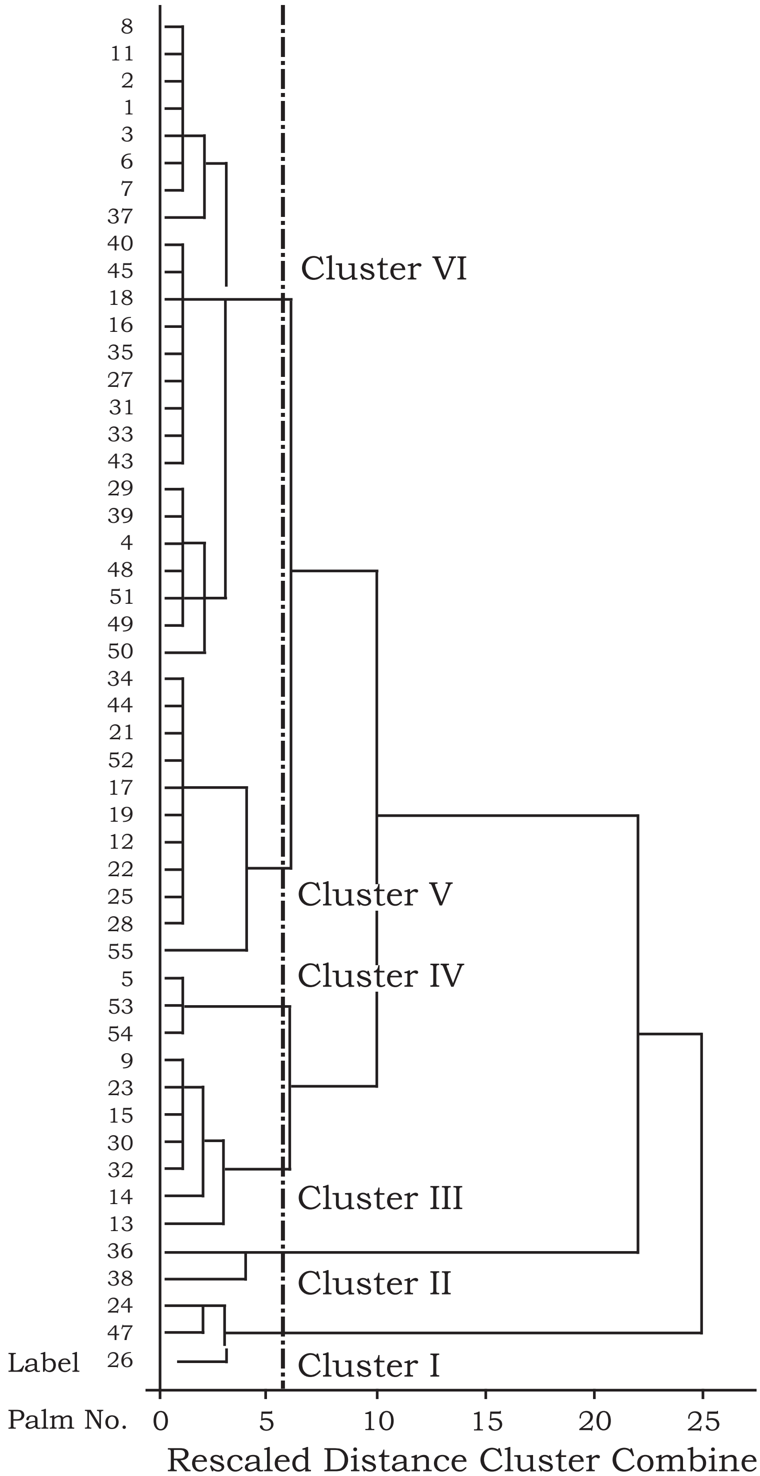

Figure 1. Dendrogram showing average linkage cluster analysis (UPGMA)

Table 8. Cluster means for 12 characters of UP makapuno coconut population

Characters	Cluster Number					
	I	II	III	IV	V	VI
Girth at bole (cm)	219.00	161.00	165.00	141.00	213.00	223.00
Girth at trunk (cm)	129.00	100.00	84.00	94.00	124.00	119.00
Length of 11 leaf scars (cm)	68.00	88.00	91.00	78.00	74.00	91.00
Fruit wt (g)	760.67	1783.00	1166.67	2133.34	1433.34	1616.67
Nut wt. (g)	450.00	1166.67	750.00	1000.00	1100.00	700.00
Husk wt. (g)	310.67	616.33	416.67	1133.34	333.34	916.67
Leaf number	28.00	29.00	22.00	24.00	34.00	32.00
Leaf petiole diameter (cm)	112.00	115.00	114.00	112.00	115.00	108.50
Fruit polar diameter (cm)	18.00	17.00	16.00	26.00	18.00	19.00
Fruit equatorial length (cm)	13.00	20.00	18.00	17.00	16.00	18.00
Nut polar length (cm)	9.20	10.00	9.50	11.50	11.00	10.20
Nut equatorial length (cm)	9.00	12.00	11.00	11.00	12.00	12.00

Fruit= fruit including husk; Nut = fruit without husk

Principal Component Analysis

Two linear combinations were identified namely first Principal Component (Prin1) and second Principal Component (Prin2) accounting for 99% of the observed variation. The rest of the components accounted for 0.01% of the variation (Table 6). Based on dendrogram, the population was distributed in six clusters on 1 to 5 scale (Figure 1). The inter-cluster distance was maximum in between clusters I and IV (1603.219) while it was minimum in between clusters V and VI (146.21). The intra-cluster distance was maximum in cluster I (163.58) and minimum in cluster II (110.19). Intra-cluster distances were identical in cluster IV and V (Table 7). Fruit and nut weight in all the clusters were found to contribute maximum diversities (Table 8). Cluster I had the smallest fruit, nut and husk weight, while cluster IV had the highest values for those cluster. Cluster II had the highest nut weight, leaf parameters but fruit size hardly contributed in observed diversity.

The population was heterogeneous in nature except for the makapuno character. It is thus concluded that high yield of makapuno was the evidence of self-pollination as well as cross-pollination among the makapuno palms. Further investigations can be done for evaluation of interaction of nut age and genotypes if any by selecting genetically uniform palms and harvesting of nuts at different ages on the basis of date of artificial pollination and fruit set. Since, makapuno is the effect of pollen parent, studies on the effect of pollen source on the in vitro germination is essential to identify inbreeding or heterosis.

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