

Development of Self-pollinating and Early-bearing Philippine Makapuno (*Cocos nucifera* L.) Hybrids

Tessie C. Nuñez and Edwin T. Ocoy

*National Coconut Research Center-Visayas (NCRC-V), Visayas State University,
Baybay City, Leyte, 6521-A Philippines*

ABSTRACT

The makapuno-bearing character found only in the cross-pollinated Laguna Tall coconut variety and the self-pollinating character of dwarf coconuts were combined into new makapuno genotypes through hybridization to limit cross pollination which is a problem among tall makapuno palms. F1 hybrids between Coconiño (CÑO) and tall Makapuno (TMAC) and Tacunan (TAC) x TMAC were early-bearing with high intraspadix overlapping percentages of 85.7% and 70.9%, respectively. Mean makapuno yield was 26.8% in CÑO x TMAC and 28.2% in TAC x TMAC indicating the presence of high degree of self-pollination and the dominance of the gene for the trait. *In vitro* grown homozygous F2 makapuno palms started flowering at 24 months old. F2 CÑO x TMAC (VMAC 1) had a mean yield of 100% makapuno, 49 to 132 nuts/palm/year and nuts weighing 332 to 750g/dehusked nut. F2 MRD x TMAC (VMAC 2) yielded a mean of 97% makapuno, 63 to 163 nuts/palm/year and nuts weighing 288 to 1,180g each. F2 TAC x TMAC (VMAC5) had 95% makapuno, nuts weighing 736g to 1,975g/ dehusked nut. High makapuno yield of the F2 hybrids affirmed the dominance of the gene for self pollination in coconut. Ten month-old nuts of the F2s had at least 18 mm thick meat and Lauric acid (C12) content ranging from 47.10% to 48.60%. The three F2 hybrids were registered with the National Seed Industry Council (NSIC) in 2008 as the first homozygous makapuno hybrids in the Philippines.

Key Words: soft-endospermed coconut, intraspadix overlapping, precocity, Lauric acid

INTRODUCTION

Makapuno (*Cocos nucifera* L.) is the most economically important soft-endospermed coconut in the Philippines where it is commonly processed into food products. The Coconut Market Information Center (CMIC) listed makapuno among the ten top ranking non-traditional export products of the country in 2010. Foreign markets for makapuno show increasing demand (Agustin, 1997) but supply falls short since the 1990s (Postharvest Systems and Management Services, 1999) due to very limited makapuno stand which was reported to be 4,226 palms throughout the country (Carpio, 1997). Current assessment estimates market deficit at 3.99 million kilos of makapuno meat (Department of Agriculture, 2011). In the Philippines, 4.061 million kilos of makapuno meat are needed by the ice cream industry while the global ice cream industry uses an estimated 1.5 billion nuts.

Majority of the accounted makapuno were the naturally existing heterozygous palms, called 'Kabuwig', found sporadically among the Laguna Tall coconut populations. 'Kabuwig' yields 2-21% makapuno (Rillo, 1997) and bears flowers at five to seven years old being tall (The Coconut Committee, 1993). These scarce resources might also be dwindling fast due to cutting of old coconut palms.

Efforts to increase makapuno production are hampered by lack of sufficient sources of makapuno planting materials, long juvenile phase and low yield of naturally existing makapuno palms. The problem of low yield was significantly reduced when Dr. E. V. de Guzman of UP Los Baños developed a means of growing *in vitro* the homozygous or true-to-type makapuno embryos which do not germinate in the nut (De Guzman, et al., 1971). These *in vitro* grown palms were supposed to yield 100% makapuno. However, the reported yield of the first homozygous makapuno palms was 67% (The Coconut Committee, 1993) and 70% (Philippine Coconut Research and Development Foundation, 1985). Yield reduction was apparently due to cross pollination with normal coconuts surrounding the makapuno-bearing palms.

To improve yield, vulnerability to pollen contamination must be reduced through the incorporation of the self-pollinating gene/character found among the dwarf coconuts into the makapuno-bearing palms. In addition, dwarf coconuts are also precocious and may impart earliness of bearing to the hybrids. Described herein are the growth, flowering and yield characteristics of the first Philippine hybrids with the combined self-pollinating and makapuno-bearing characters developed at the National Coconut Research Center-Visayas, Visayas State University in Baybay City, Leyte, Philippines.

MATERIALS AND METHODS

Development and Evaluation of First Filial Generation Dwarf x Tall Makapuno Hybrids

Three self-pollinating dwarf (D) coconut cultivars namely, Coconiño (CÑO), Malayan Red Dwarf (MRD) and Tacunan (TAC) were used as female parents to produce three first filial generation (F1) makapuno hybrids (Fig. 1). Makapuno pollen was obtained from embryo-cultured tall makapuno (TMAC) of the Department of Horticulture, University of the Philippines at Los Baños (UPLB). Hybridization work was done at the Visayas State College of Agriculture (now Visayas State University) following the controlled-hand-pollination method for coconut (Santos, et al., 1996). The resulting heterozygous F1s were field-planted in a replicated set up, except MRD x TMAC which had very few surviving seedlings, to characterize their growth, flowering, and makapuno yield.

In vitro Culture of Second Filial Generation Homozygous Makapuno Hybrids

Ten-month old second filial generation (F2) makapuno nuts were harvested from the heterozygous F1 makapuno hybrid palms. Embryos were extracted from the makapuno nuts and grown *in vitro* following standard procedures (Rillo, 2000) using Y3 culture medium (Eeuwens, 1976) during the early part of the study. The culture medium was later modified using a different vitamin component for better growth and development of the makapuno hybrid embryos (Nuñez, 2007). Developing F2 homozygous makapuno hybrid seedlings were grown in the laboratory for at least six months before they were potted and kept in a screen house to harden for three to six months.

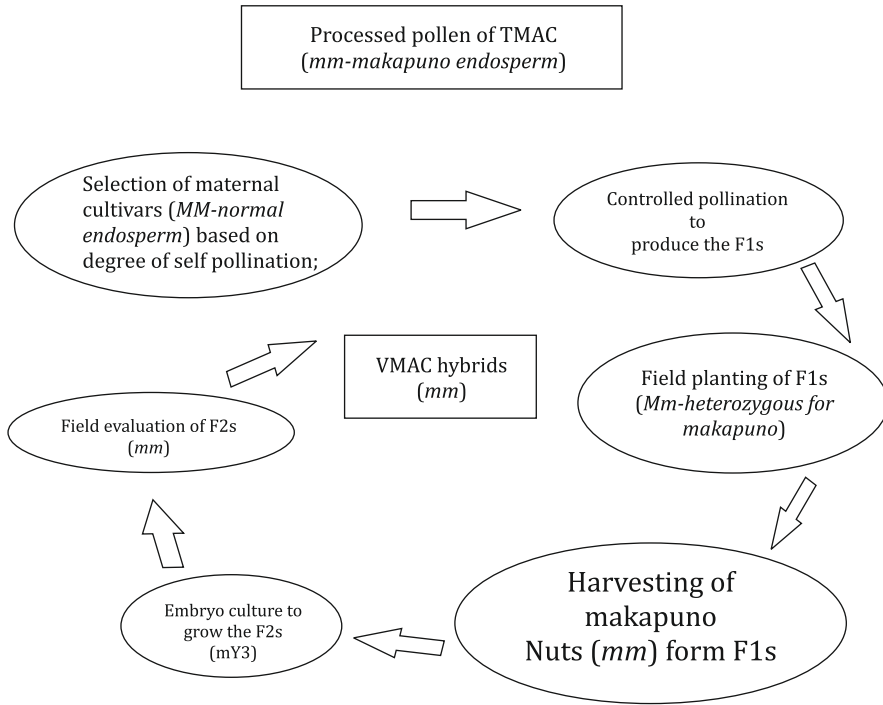


Figure 1. Process of development of the Makapuno hybrids

Evaluation of F2 Homozygous Makapuno Hybrids

Fully developed F2 homozygous makapuno seedlings of CÑO x TMAC (VMAC 1), MRD x TMAC (VMAC 2), and TAC x TMAC (VMAC 5) were field-planted in a replicated trial to characterize their growth, flowering and yield. Field planting was done following the triangular planting scheme with 8.5m distance between hills. Palms were applied with the recommended amounts of inorganic fertilizers (The Coconut Committee, 1993).

Morphology of palms was categorized as dwarf when they have straight and thin stems. Those with boles and medium to big stems were classified as D x T hybrids in growth morphology. Age at initial flowering of palms and the degree of intraspadix overlapping of the male and female phases were also determined as indications of earliness of bearing and self pollination, respectively.

Degree of intraspadix overlapping was measured as percent of the period of stigmatic receptivity overlapped by the period of pollen dehiscence in an inflorescence. High degree of intraspadix overlapping indicates high degree of self-pollination.

Yield was accounted as number and % makapuno nuts per bunch per harvest and presented as number and % makapuno nuts per palm per year. Size of makapuno nuts was measured as weight of dehusked nut.

Meat thickness and fatty acid profile of 10 month-old makapuno nuts were also determined because of their importance in the use of makapuno as food. The Biochemistry Section of the Philippine Coconut Research and Development Foundation, Inc. (PCRDF) in Manila did the fatty acid analysis.

RESULTS AND DISCUSSION

Growth, flowering and yield characteristics of D x T F1 makapuno hybrids

Trunks of most F1 palms of CÑO x MAC and TAC x MAC were developed during the early bearing stage. At 5-6 years after planting, trunks were less than a meter in height (Table 1).

Table 1. Growth, flowering, and yield of F1 D x T makapuno hybrids

Character	F1 CÑO x MAC	F1 TAC x MAC
Trunk height (m) at 5-6 years old	0.57-0.69	0.38-0.46
Annual mean leaf production at early maturity (5-10 years old)	13-16	10-17
Age at initial flowering (months)	39 - 66	52 - 67
Mean intraspadix overlapping (%) ^{1/}	85.7	70.9
Makapuno nut/palm/year	31-99	31-157
Makapuno Yield (%) /year	17.7-44.0 (Mean - 26.8)	16.0-47.0 (Mean - 28.2)
Dehusked ^{1/} nut weight (g)	351-1,565	711-1,832

^{1/} Husk was removed

Leaf production at early maturity showed fair to good growth of the F1 CÑO x TMAC and TAC x TMAC with at least 10 leaves produced per year starting at five years after planting. A coconut palm normally bears one leaf per month at maturity. The number of leaves produced also indicates the maximum number of bunches that the palms can yield in a year since each leaf carries a flower bud.

F1 CÑO x TMAC palms bore flowers at 39 to 66 months after planting while TAC x TMAC palms started to flower at 52 to 67 months after planting. Tall coconuts are known to flower at 5-7 years after planting while the dwarfs, like CÑO and TAC, normally bear flowers at 3-4 years from planting (Banzon & Velasco, 1982; The Coconut Committee, 1993). Earliness of bearing of the F1 hybrids was due to the influence of the dwarf maternal parents.

Both hybrids showed considerable degree of intraspadix overlapping which is an indication of the extent of self pollination in them. CÑO x TMAC had a mean intraspadix overlapping of 85.7% while TAC x TMAC had 70.9%. TMAC, which was the male parent of the hybrids, is cross-pollinated being tall while the maternal parents, CÑO and TAC, are self pollinated. The high intraspadix overlapping percentages of the F1 hybrids implied that the gene for self pollination in coconut is dominant over the gene for cross pollination.

Makapuno nut production per palm per year showed wide range of values in both hybrids because they are heterozygous and possibly partly due to environmental influence. Percent makapuno yield was high with a mean of 26.89% in CÑO x TMAC, and a mean of 28.2% in TAC x TMAC showing the presence of high degree of self pollination among the palms. Theoretically, the F1 hybrids would yield 25% makapuno when there is 100% self pollination. Higher makapuno yield of the F1 hybrids also suggested that blocked planting might have contributed to higher makapuno yield of the palms. Since almost all palms were surrounded by heterozygous palms, makapuno pollen from other palms could have pollinated the flowers of another palm. This observation shows the importance of planting makapuno palms in block as recommended by The Coconut Committee (1993) to improve yield.

Makapuno nuts of the F1 hybrids also varied in shape and size. Shape of nuts generally resembled the shape of the maternal parent (Fig. 2). Nut size appeared to be influenced by the genotype of the parents and the number of nuts produced by the palm. Production of more nuts reduced nut size while bigger nuts were observed when fruits were few. CÑO x MAC, whose

maternal parent is small-seeded, produced small to big nuts with weights ranging from 351 to 1,565g each when dehusked. TAC x MAC, whose maternal parent is medium-seeded, bore medium to big nuts weighing 711g to 1,832g each when dehusked. Wide variation in size of nuts might also be partly due to segregation of the genes for this trait during gamete formation of the F1s which were heterozygous possibly for almost all traits. In addition, environmental effects during the almost 10-year evaluation period could have also contributed to the variations in nut size.

Growth morphology, flowering and yield of the F2 homozygous makapuno hybrids

VMAC1 had the dwarf and D x T hybrid type morphologies. It had the shortest and smallest trunks, in terms of girth size (Fig. 3a). Initial flowering in VMAC1 started at 24 to 37 months after planting (Table 2). Intrapadix overlapping was high with a mean of 93.90%. VMAC2 and VMAC5 generally had the D x T hybrid type morphology (Figs. 3b and 3c). They also had taller and bigger trunks than VMAC1. Age at initial flowering ranged from 24 to 36 months from planting in VMAC2 and 34 to 48 months from planting in VMAC5. Mean intraspadix overlapping was 60.30% in the former and 67.17% in the latter. F2 makapuno nuts resemble the shape of their maternal parents except for thicker and soft meat (Fig. 4).

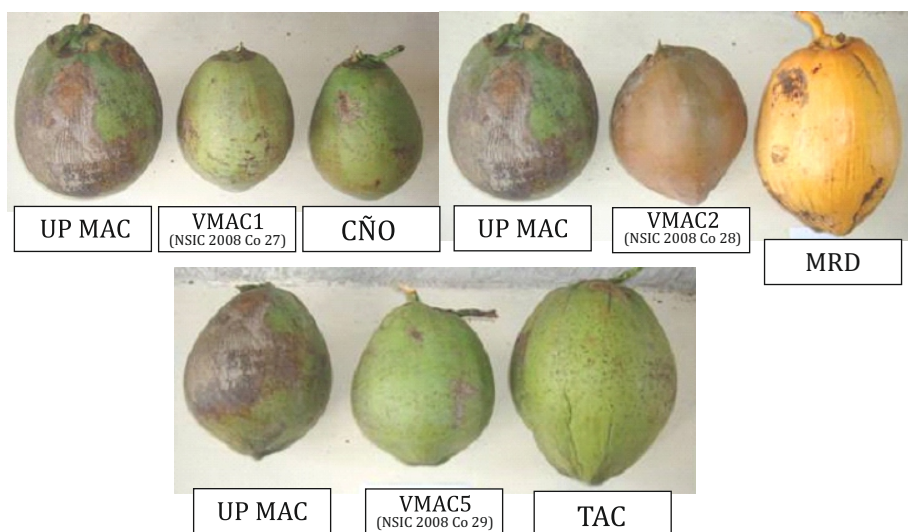


Figure 2. Nut shapes of the parental cultivars and the VMAC hybrids

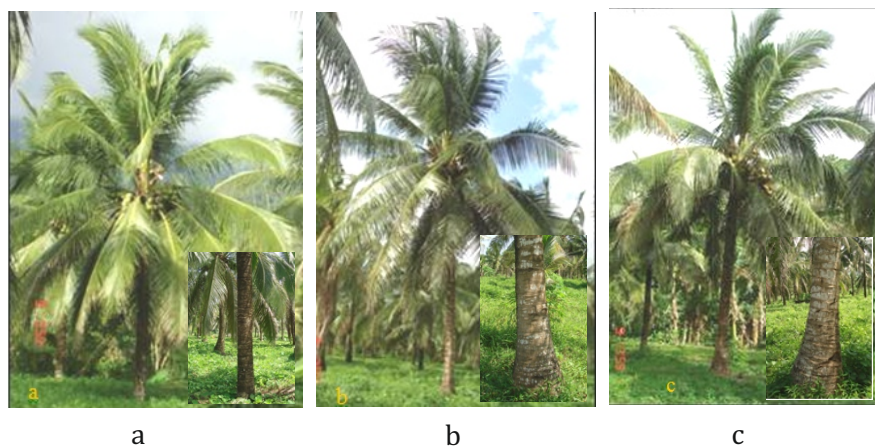


Figure 3: Characteristic morphology of NSIC-registered makapuno hybrids:
a) VMAC1, b) VMAC2 and c) VMAC5



Figure 4. Split F2 makapuno nuts

Table 2. Flowering characteristics of F2 homozygous makapuno hybrids

Hybrid	Age at initial flowering (months)	Mean Intraspadix Overlapping (%)
VMAC1	24-37.	93.90
VMAC2	24-36	60.30
VMAC5	34-48	67.16

Among the homozygous F2s, VMAC 1 had the highest mean makapuno yield of 100% (Table 3). VMAC2 and VMAC5 had 97% and 95% makapuno yield, respectively. Number and size of makapuno nuts produced by the three hybrids were very variable because of their genotypes and environmental influence. VMAC1 produced small to medium nuts. VMAC2 had small to big while VMAC5 produced medium to big nuts (Table 3). All hybrids had a mean meat thickness of about 18 mm.

Table 3. Yield characteristics of F2 homozygous makapuno hybrids

Hybrid Cross	Number of makapuno nuts/palm/year	% makapuno	Dehusked nut Weight (g)	Meat thickness (mm)*
VMAC1	49-132	100	332-750	18.5
VMAC2	63-163	97	288-1,180	18.0
VMAC5	36-126	95	736-1,975	18.4

* Ten-month old nuts

A fatty acid profile analysis was likewise done on dried meat of homozygous makapuno hybrids. Results showed that Lauric acid (C12) was the main component of the oil at 48.60% in VMAC1, 47.87% in VMAC2 and 47.10% in VMAC5 (Table 4). Similar results were reported by Gunathilake, et al., (2009) in their analysis of Dikiri coconut in Sri Lanka. Dikiri coconut is very similar to the Phillipine makapuno.

Table 4. Fatty acid profile of ten-month old F2 makapuno hybrid nuts

Fatty Acid	Relative Mass Percentage (%)		
	VMAC 1	VMAC 2	VMAC 5
Caproic (C6)	0.73	0.75	0.73
Caprylic (C8)	7.81	8.06	7.79
Capric (C10)	5.41	5.66	5.44
Lauric (C12)*	48.60	47.87	47.10
Myristic (C14)	19.62	18.74	19.59
Palmitic (C16)	8.89	8.77	9.27
Stearic (C18:0)	3.12	3.22	3.79
Oleic (C18:1)	4.89	5.95	5.39
Linoleic (C18:2)	0.92	0.97	0.89

* Lauric acid contents: 50.06% in mature tall makapuno and 45.9%-50.3% in mature ordinary coconut (Canapi, et al., 2005)

Registration of the homozygous makapuno hybrids

VMAC1, VMAC2 and VMAC5 were registered with the National Seed Industry Council of the Philippines in 2008 as NSIC 2008 Co 27, NSIC 2008 Co 28, and NSIC 2008 Co 29, respectively, the first homozygous makapuno hybrids in the country.

CONCLUSION

The self-pollinating character, which apparently was controlled by a dominant gene, was incorporated in the first reported Philippine makapuno hybrids described in this paper, together with earliness of bearing. The tendency of the said hybrids to self pollinate is believed to significantly reduce vulnerability to unwanted pollination that could reduce makapuno yield although yield might still be reduced by insect pollination. Availability of early-bearing makapuno palms may also contribute significantly to faster production of homozygous makapuno planting materials to hasten the growth of makapuno production in the country.

LITERATURE CITED

- AGUSTIN Y. T. V. 1997. Market Development and opportunities for Makapuno. In Makapuno Development and Market Trends. Seminar Workshop Proceedings. pp.13-19.
- BANZON, J. A. and J. R. VELASCO. 1982. Coconut Production and Utilization. PCRDF, Inc. Amber Avenue, Pasig, Metro Manila, Philippines. p.136
- CANAPI, E. C., Y. T. V. AGUSTIN, E. A. MORO, E. PEDROSA, JR., M. LUZ and J. BENDAN~ O. 2005. Bailey's Industrial Oil and Fat Products, Sixth Edition, Six Volume Set . Fereidoon Shahidi (Ed.). John Wiley & Sons, Inc. p. 138

- CMIC. 2010. Performance of Philippine Top Non-Traditional Coco Exports in February 2010. Coconut Market Information Center. Retrieved April 13, 2011 from <http://www.coconutmic.com/en/market-information/export-and-import/102-performance-of-philippines-top-non-traditional-coco-exports-in-february-2010>
- DE GUZMAN, E. V.; A. G. DEL ROSARIO and E. C. EUSEBIO. 1971. The growth and development of coconut makapuno embryo. *Phil. Agric.* **53(10)**:556-579.
- Department of Agriculture, 2011. Seedling mass production facility put up to aid country to become world's biggest makapuno producer. Department of Agriculture Official Gazzete.
- EEUWENS, C. J. 1976. Mineral requirements for growth and callus initiation of tissue explants excised from mature coconut palm (*Cocos nucifera*) and cultured *in vitro*, *Physiol. Plant.* **36**:23-28
- GUNATHILAKE, K.D.P.P., C. THILAKAHEWA and A.A.N. KUMARA. 2009. Nutritional composition of Dikiri Coconut. *CORD* **25 (2)**:56-62
- NUÑEZ, T. C. 2007. Comparative suitability of two culture media to the *in vitro* growth of embryos of three coconut types. *Ann. Trop. Res.* **29**:79-90
- PCDRF. 1985. Makapuno embryo culture. In PCRDF- the first decade 1975-1985. Quezon City.
- PSMSI. 1999. Feasibility Study on Embryo -Cultured Makapuno Production. Prepared by Postharvest Systems and Management Services for TAPI-DOST, PCARRD and PCA.
- RILLO, E. P. 1997. Makapuno Embryo Culture Technology. In Makapuno Development and Market Trends. Seminar Workshop Proceedings. pp. 28-45.
- RILLO, E. P. 2000. Embryo Cultured Makapuno: from the Laboratory to the Farm. In Selected Topics on Current Trends and Prospects in Enhancing the Coconut Industry. Proceedings of the Coconut Week Symposium 2000, August 29, 2000 PCA, Diliman, Quezon City, Philippines pp.6-20

SANTOS, G.A, P.A. BATUGAL, A. OTHMAN, L. BOUDOUIN, and J.P. LABOUISSSE. 1996. Manual on Standardized Research Techniques in Coconut Breeding. COGENT, IPGRI.

THE COCONUT COMMITTEE. 1993. The Philippine Recommends for Coconut. Los Banos, Laguna: PCARRD, PARRFI and PCRDF. pp. 30-31