AJCS 11(12):1508-1519 (2017) doi: 10.21475/ajcs.17.11.12.pne523



Characterization, pollen behavior and propagation of five selected *Hibiscus* hybrids (*Hibiscus rosa-sinensis* Linn.)

Alangelico O. San Pascual¹, Pablito M. Magdalita^{1,2*}, Norma G. Medina² and Bryan V. Apacionado²

¹Institute of Plant Breeding, College of Agriculture and Food Science, University of the Philippines Los Baños, College, Laguna, 4031, Philippines ²Institute of Crop Science, College of Agriculture and Food Science, University of the Philippines Los Baños

²Institute of Crop Science, College of Agriculture and Food Science, University of the Philippines Los Baños, College, Laguna, 4031, Philippines

*Corresponding author: pabsmagdalita@gmail.com

Abstract

Breeding and characterization of Hibiscus hybrids in the Philippines was conducted to improve the local varieties for them to have unique flower colors and forms by introducing genes from foreign varieties. The Hibiscus hybrids are important ornamental genetic materials being used to honor outstanding Filipina achievers. The study aimed to characterize 5 new selected hybrids, test their pollen fertility and evaluate their graft compatibility with 2 known rootstock varieties. The important characteristics of the 5 selected hybrids are as follows: The flower of hybrid H. rosa-sinensis 'Accession 20' x H. rosa-sinensis 'Gelia Castillo' is solferino purple or red purple (RHCC 65B) with a ruby red (RHCC 59A) eye surrounded by gravish violet (RHCC N77C) halo, while the bloom of hybrid H. rosa-sinensis 'Tandang Sora' x H. rosa-sinensis 'Connie S. Angeles' is tuscan yellow (RHCC 162C) with ruby red (RHCC 59A) eye surrounded by reddish white (RHCC N1555A) halo. The petal of hybrid H. rosa-sinensis 'Tarantella' x H. rosa-sinensis 'Golden Dubloom' is rose red (RHCC 58B) surrounded with yellow orange (RHCC 20A) edging, while the eye is dark red rose (RHCC 61B). The flower of hybrid H. rosa-sinensis 'Perla Santos-Ocampo' x H. rosa-sinensis 'Loren B. Legarda', is spanish orange (RHCC 26B) with lemon yellow (RHCC 13B) eye and dark orange markings (RHCC N 25B) radiating from the eye going to the petals. In addition, the petal color of hybrid H. rosa-sinensis 'Loren B. Legarda' x H. rosa-sinensis 'Tarantella' is a combination of primrose yellow (RHCC4B) and neyron rose (RHCC 58C). Correlation analysis was conducted for the different floral and leaf traits such as corolla length, width, bloom length, receptacle diameter, style length, calyx lobe width, sepal length, style length, ovary width, petiole length, leaf length and width. Very high positive correlation (r = 0.9428) was detected on corolla length and bloom length. High correlation was observed on bloom length and corolla width (r=0.8663), corolla width and corolla length (r= 0.8727), style length and bloom length(r=0.7136), receptacle diameter and calyx lobe width (r=0.7861). Pollen viability of the 5 different Hibiscus hybrids when used as male parents in hybridization, as shown by potassium iodide structural staining was 61.62 to 70.0%, but significant differences on pollen viability were not detected among the different hybrids. However, significant differences between different sucrose concentrations added to the Brewbaker and Kwack's (BK) medium for pollen germination test of H. rosasinensis 'Accession 20' x H. rosa-sinensis 'Gelia Castillo' were detected. Ten percent (v/v) sucrose as additive to BK medium was the optimum concentration for pollen germination at 70%. Good pollen germination, a prerequisite for pollen tube growth in the stigma going to the ovary, hence effective fertilization and development of the zygote and growth of the embryo in the hybrid seed, is necessary for successful hybridization. Furthermore, evaluation of the graft compatibility of the 5 Hibiscus hybrids to known rootstock varieties was conducted as a prerequisite for efficient asexual propagation. Significant difference ($p \le 0.05$) was detected between rootstock varieties *H. rosa-sinensis* 'Reddy or Not' and *H. rosa-sinensis* 'Wilcox' when side-grafted with the hybrid *H.* rosa-sinensis 'Accession 20' x H. rosa-sinensis 'Gelia Castillo'. Rootstock variety H. rosa-sinensis 'Wilcox' promoted better grafting success by 65% when used for side-grafting H. rosa-sinensis 'Accession 20' x H. rosa-sinensis 'Gelia Castillo' compared to rootstock variety H. rosa-sinensis H. rosa-sinensis 'Reddy or Not'. However, no significant difference in the grafting success was detected between H. rosa-sinensis 'Wilcox' and H. rosa-sinensis 'Reddy or Not' when side-grafted with the other Hibiscus hybrids. Overall, it is recommended that the 5 Hibiscus hybrids be used as potted and landscaping materials, and as parents for hybridization with other varieties, while the fertile pollen could be utilized for cross-pollination with other varieties to produce another set of Hibiscus hybrids.

Keywords: Brewbaker and Kwack's, gumamela, *Hibiscus rosa-sinensis*, ornamental breeding, phenotypic characterization, pollen fertility.

Abbreviations: ANOVA _analysis of variance; BK_ Brewbaker and Kwack's; CRD _Completely Randomized Design; LSD _ least significant difference; I_2KI_p potassium iodide; RHCC_ Royal Horticutural Colour Chart; RHS _Royal Horticultural Society; nd_ no date; var._ variety; v/v _volume over volume.

Introduction

Hibiscus rosa-sinensis L., or gumamela, is a native of tropical and sub-tropical regions of the world. It belongs to the family Malvaceae consisting of 400 species (Pancho and Gruèzo, 2006). The *Hibiscus* is a diploid species with a basic

chromosome number, x = 7, 8 to 39 (Purseglove, 1987). It was believed that the Chinese traders introduced the *Hibiscus* to the Philippines long before the coming of the Spaniards (Cinco, 2013). On the other hand, the American colonizers

who came to the country in the 1890's introduced new forms and cultivars. The beauty of the *Hibiscus* makes it one of the most widely cultivated flowers in brilliant hues of red, orange, or purplish-red, yellow, white, purple, pink and several other color combinations that are either single or double petals. From a flower bud, the flower emerges in less than 18 hours usually in the morning during sunny days. The plant has short-lived flowers, usually lasting for 1 to 2 days, but produces continuing blooms throughout the year. Under natural conditions, the *Hibiscus* rarely produces fruit and seeds (Howie 1980).

In the Philippines, the Hibiscus has become a popular ornamental plant with attractive flowers commonly used for adorning many Filipino home gardens, facades, golf courses and public places such as schools, parks and resorts. It is also used either as a potted plant, fence or hedge plant and landscape shrub (Magdalita et al., 2009; Magdalita and Pimentel, 2010). Large numbers of plant sasexually propagated hybrids were supplied semi-commercially to various Hibiscus stakeholders for adoring job centers and business establishments and for edible landscaping purposes (Magdalita and Pimentel, 2013). It is also used for food, feed, industrial and medicinal preparations. For instance, it is used to treat epilepsy, leprosy and diabetes (Kumar and Singh, 2012; Cinco, 2013). The fresh flowers are used as food coloring and as component of vegetable salad, while the dried flowers are considered a special delicacy in Mexico.

The leaves are used as herbal tea which is gaining popularity especially in Asia because of its perceived association with longevity. The natives of southern India use the red Hibiscus as hair shampoo for the remedy of hair-fall and dandruff by inducing the hair follicles to make protective oils (Rummel, 2005). The flowers contain vitamins, flavonoids, ascorbic acid, niacin, riboflavin, thiamine and cyanadine diglucoside (Pekamwar et al., 2013). Furthermore, the flowers contain the compounds called flavonol quercetin and anthocyanin cyanidin (Rummel, 2005). The white or cream colored Hibiscus varieties contain high levels of quercetin, while the dark magenta-red varieties are very rich in cyanidin. In experimental mice, the application of flower extracts for 20 weeks significantly reduced the number of tumors per mouse and delayed the appearance of the first tumor when mice were pre-treated with the extract (Rummel, 2005). Due to its high flavonoid and terpenoid content, it shows significant antioxidant and anticancer activities (Divya et al., 2013).

In the Dutch Indies, the midwives use the mucilage of the *Hibiscus* flower to induce mother's delivery of the baby during labor. The juice of the leaves along with that of *Vernonia cinerea* is also used by midwives to stimulate the expulsion of the afterbirth. The red *Hibiscus* flowers are used to regulate menstruation, and are purgative. In Chinese medicine, the crude extract of the bloom is used effectively for decreasing the level of glucose and cholesterol in humans (Mishra et al., 2009).

The production of different hybrids for potted plants is a very lucrative income generating business due to high demand for landscaping materials (Magdalita et al., 2016). Due to their promising demand, *Hibiscus* breeding has been conducted at the Institute of Plant Breeding, College of Agriculture and Food Science, University of the Philippines Los Baños. This breeding work have resulted to the development and release of different hybrids belonging to the following series: a) Centennial Series consisting of 9 hybrids named after the heroines of the 1898 Philippine Revolution against the Spaniards (Pimentel, 1999), b) Millenium Series consisting of 7 hybrids named after women scientists of University of the Philippines Los Baños who contributed to the advancement of Philippine Agriculture, and c) Celebrity Star Series consisting of 5 hybrids named after accomplished celebrities in the Philippine Cinema (Magdalita and Pimentel, 2010). To mark the University of the Philippines Centenary in 2008, new hybrids were also released belonging to the Oblation Series consisting of 6 hybrids named after outstanding University of the Philippines alumnae who have assumed the highest position in the academic, scientific and professional institutions and organizations where they served (Magdalita et al., 2009). In addition, the Women in Public Service Series consisting of 12 hybrids named after outstanding public servants, Women in Education Series (1 hybrid), Women in Science Series (4 hybrids) and Women Saint Series (1 hybrid) were also released (Magdalita et al., 2011; Magdalita and Pimentel., 2013).

In the ornamental industry, the production of potted live ornamental plants for direct sale is a very good income generating business for the local market. Studies have shown that the biggest market for Philippine export of potted and live plants is Korea, where they absorbed 65% of the total volume valued at 9.7 million pesos. In addition, the highest export earnings of the country from ornamental plants came from potted and live plants at an average of 23.8 million pesos annually for a period of 9 years (Naranja, 2007; Pabuyon, 2001). This indicates a very bright future for the potted plant industry, where the new Hibiscus hybrids could be a component that can further increase the revenue of the country. Hence, the continuous breeding of new Hibiscus hybrid varieties is necessary. In addition, these hybrids need to be propagated to mass produce them for dissemination to various stakeholders. Since they are new hybrids, their graft compatibility with known rootstocks need to be evaluated.

The variability in flower colors and form, plant growth habit and *in situ* resistant reaction to pests and diseases of related *Hibiscus sp.*, local varieties and foreign hybrids in the *Hibiscus* germplasm were exploited for hybrid breeding by the breeders-researchers of the Institute of Plant Breeding (Pimentel, 1999; Magdalita et al., 2009). The continuous hybridization of *Hibiscus* requires identification of parents with good combining ability and those that are good pollinators. Genotypes that are good sources of pollen can be selected by testing pollen fertility. Structural staining using potassium iodide (I₂KI) and pollen germination tests using Brewbaker and Kwack's (BK) medium (1963) can be used for testing pollen fertility.

The study was conducted primarily to describe the novel traits of newly developed *Hibiscus* hybrids via cross-pollination followed by selection; hence, these hybrids become unique genetic materials in the *Hibiscus* gene pool. Hence, the study was conducted to: i) characterize the qualitative and quantitative traits of the selected hybrids, ii) evaluate their pollen fertility, and iii) evaluate the graft compatibility of the new hybrids using 2 rootstock *Hibiscus* varieties.

Results

Phenotypic characterization

Selected *H. rosa-sinensis* hybrids collectively called the 'Women in Education Series' were characterized phenotypically. Five hybrids including: *H. rosa-sinensis* 'Accession 20' x *H. rosa-sinensis* 'Gelia Castillo', *H. rosasinensis* 'Tandang Sora' x *H. rosa-sinensis* 'Connie S. Angeles', *H. rosa-sinensis* 'Loren Legarda' x *H. rosasinensis* 'Tarantella', *H. rosa-sinensis* 'Tarantella' x *H. rosasinensis* 'Golden Dubloom', and *H. rosa-sinensis* 'Perla

Table 1. The pedigree and phenotypic characteristics of the five Hibiscus hybrids

Female parent	Male parent	Bloom characteristics	Foliation	Growth habit
H. rosa- sinensis 'Accession 20'	<i>H. rosa-sinensis</i> 'Gelia Castillo'	Solferino purple (RHCC 65B) petals with ruby red (RHCC 61A) eye surrounded by grayish halo	Ovate , 91mm long, 88mm wide, simple, green (RHCC N137B), glossy and smooth	Medium height (1.37 m), erect growth habit, shrubby (0.77 m), very floriferous
H. rosa- sinensis 'Loren B. Legarda'	<i>H. rosa-sinensis</i> 'Tarantella'	Primrose yellow (RHCC 4B) petal with neyron rose (RHCC 58C) halo	Ovate, 117.3mm long and 90.8 mm wide, simple, green (RHCC N137C), glossy and smooth	Semi-dwarf in height (0.81 m), erect growth habit, moderately shrubby (0.57 m), floriferous
H. rosa- sinensis 'Tarantella'	<i>H. rosa-sinensis</i> 'Golden Doubloom'	Rose red flower with yellow orange edging and red rose eye	Ovate, 121.3 mm long and 89.9 mm wide, simple, green (RHCC N137C), glossy and smooth	Medium height (1.29 m), semi-erect growth habit, shrubby (0.68 m), floriferous
H. rosa- sinensis 'Tandang Sora'	H. rosa-siensis 'Connie S. Angeles'	Tuscan yellow petal with ruby red eye surrounded by reddish white halo radiating from the eye going to the petal	Ovate, 85.9mm long, 61.3 mm wide, simple, glossy green (RHCC 137A), smooth	Medium height (1.29 m), semi-erect growth habit, shrubby (0.86 m), floriferous
<i>H. rosa- sinensis</i> 'Perla Santos- Ocampo'	<i>H. rosa-sinensis</i> 'Loren B. Legarda'	Spanish orange petal with lemon yellow eye and dark orange halo radiating from the eye to the petal	Ovate, 107.7 mm long and 77.7 mm wide, simple, green (RHCC N137B), smooth and glossy	Tall height (2.09 m), semi- erect growth habit, shrubby (1.17 m), very floriferous

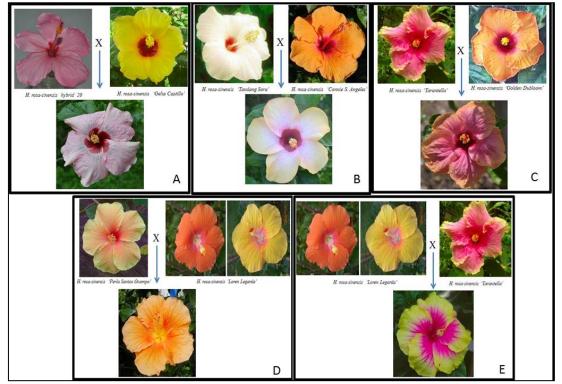


Fig 1. *H. rosa-sinensis* 'Accession 20' x Gelia Castillo' and its female, *H. rosa-sinensis* 'Accession 20' and male, *H. rosa-sinensis* 'Gelia Castillo' parents (A), *H. rosa-sinensis* 'Tandang Sora' x 'Connie S. Angeles', its female, *H. rosa-sinensis* 'Tandang Sora' and male, *H. rosa-sinensis* 'Connie S. Angeles' parent) (B), *H. rosa-sinensis* 'Tarantella' x 'Golden Doubloom', its female, *H. rosa-sinensis* 'Tarantella' and male, *H. rosa-sinensis* 'Golden Dubloom' parent) (C), *Hibiscus rosa-sinensis* 'Perla Santos-Ocampo x 'Loren B. Legarda' its female, *H. rosa-sinensis* 'Perla Santos- Ocampo' and male, *H. rosa-sinensis* 'Loren B. Legarda' parents (D), *H. rosa-sinensis* 'Loren B. Legarda' x 'Tarantella', its female, *H. rosa-sinensis* 'Loren B. Legarda' and male, *H. rosa-sinensis* 'Tarantella', its female, *H. rosa-sinensis* 'Loren B. Legarda' and male, *H. rosa-sinensis* 'Tarantella', its female, *H. rosa-sinensis* 'Loren B. Legarda' and male, *H. rosa-sinensis* 'Loren B. Legarda' x 'Tarantella', its female, *H. rosa-sinensis* 'Loren B. Legarda' and male, *H. rosa-sinensis* 'Tarantella', its female, *H. rosa-sinensis* 'Loren B. Legarda' and male, *H. rosa-sinensis* 'Loren B. Legarda' and male, *H. rosa-sinensis* 'Tarantella' parents.

 Table 2. Correlation coefficient among characters evaluated in H. rosa-sinensis hybrids.

Character combination	Correlation coefficient (r)*
Corolla length and Bloom length	0.94
Corolla width and Bloom length	0.87
Corolla width and Corolla length	0.87
Style length and Bloom length	0.71
Receptacle diameter and Calyx lobe width	0.78
Sepal length and Style length	-0.83
Ovary width and Bloom length	0.52
Ovary width and Corolla length	0.50
Ovary width and Corolla width	0.50
Style length and Bloom length	0.66
Petiole length and Receptacle diameter	0.54
Leaf width and leaf length	0.65
Petiole length and leaf width	0.61

*Character combination included were found significant at 5% level of significance

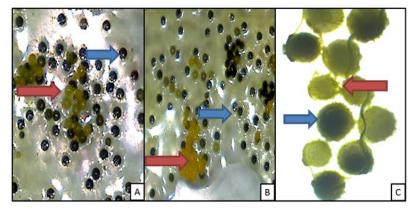
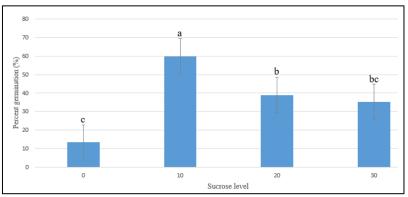


Fig 2. Pollen grains of *H. rosa-sinensis* 'Perla Santos-Ocampo x *H. rosa-sinensis* Loren B. Legarda' (A) and *H. rosa-sinensis* 'Loren B. Legarda' x 'Tarantella' (B and C) subjected to potassium iodide structural staining. Blue arrows indicate fully stained pollen while red arrows indicate partially and unstained pollen grains.

Table 3. Percent pollen fertility of different *H. rosa-sinensis* hybrids obtained using I₂KI structural staining technique.

Selected hybrids	% Viability*
Hibiscus rosa-sinensis 'Accesion 20' x H. rosa-sinensis 'Gelia Castillo'	70.00 a
H. rosa-sinensis 'Tarantella' x H. rosa-sinensis 'Golden Doubloom'	67.82 a
H. rosa-sinensis 'Tandang Sora' x H. rosa-sinensis 'Connie S. Angeles'	55.48 a
H. rosa-sinensis 'Perla Santos-Ocampo x H. rosa-sinensis 'Loren B. Legarda'	67.40 a
H. rosa-sinensis 'Loren B. Legarda' x H. rosa-sinensis 'Tarantella'	61.62 a

*Values with the same letters are not significantly different at 5% LSD



Bars with different letters are significantly different at $\alpha = 5\%$

Fig 3. Percent pollen germination of *H. rosa-sinensis* 'Accession 20' x *H. rosa-sinensis* 'Gelia Castillo' on Brewbaker and Kwack's medium supplemented with varying sucrose concentrations.

Table 4. Qualitative and Quantitative traits used to characterize the five *Hibiscus* hybrids.

Qualitative Traits	Quantitative Traits
Leaf margin	Leaf length
Leaf form and outline	Leaf width
Leaf arrangement	Petiole length
Leaf parts and types	Angle of display
Leaf apices	Corolla length
Leaf bases	Corolla width
Leaf attachments	Ovary width
Adaxial leaf color	Ovary length
Abaxial leaf color	Receptacle width
Bloom type	Pedicel length
Inflorescence type	Sepal length
Stigma type	Stigma length
Style type	Stigma width
Calyx color	Style length
Corolla color	Plant height
	Plant width

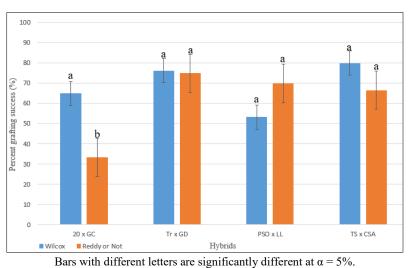


Fig 4. Percent grafting success of selected *H. rosa-sinensis* hybrids as scion, side grafted to *H. rosa-sinensis* 'Wilcox' and *H. rosa-sinensis* 'Reddy or Not' rootstocks.



Fig 5. Rootstock varieties used in the graft compatibility experiment, *H. rosa-sinensis* 'Reddy or Not' (A), *H. rosa-sinensis* 'Wilcox' (B), successfully grafted plants of different hybrids grafted to rootstocks of *H. rosa-sinensis* 'Wilcox' (C) and, *H. rosa-sinensis* 'Reddy or Not' (D).

Santos-Ocampo' x *H. rosa-sinensis* 'Loren Legarda' comprised the series. The detailed narrative descriptions of the 5 new *Hibiscus* hybrids were presented below, while the pedigree or parents of the hybrids and their important phenotypic characters were summarized in Table 1.

Hibiscus rosa-sinensis 'Accession 20' *x H. rosa-sinensis* 'Gelia Castillo'

Fig. 1a shows the hybrid between *H. rosa-sinensis* 'Accession 20' x *H. rosa-sinensis* 'Gelia Castillo'. It is shrubby with an erect growth habit, medium height at 1.37 m and a width of 0.77 m. The hybrid is a fast grower on its own root.

The leaves are ovate, with crenate margin, reniform base, apiculate apices and an alternate petiole arrangement. They are green (RHCC N137B), 91 mm long and 85 mm wide. The petiole is 22.3 mm long.

Flowers are funnel-shaped, solitary and borne in a slender stalk. The pedicel is yellow-green (RHCC 146C) and 31.1 mm long. The bracts are involucral. The bloom is simple and regular, 125.8 mm in diameter. The bloom lasts for a day only, but flowers bloom each day. Corollas are 132.5 mm long and 121.4 mm wide. The corolla is solferino purple (RHCC 65B) with a ruby red (RHCC 59A) eye surrounded by a grayish violet (RHCC N77C) halo (Fig. 1). The calyx is obovate, thick, imbricate, glabrous and green (RHCC 146B). Calyx lobes are 15.6mm long and 8.79 mm wide. The flower's angle of display on the plant is 89.4°.

The staminal tube is 59 mm long. Five capitate orange (RHCC 34A) stigmas are 2.02 mm long and 0.92 mm wide. The androecial type is monadelphous. Sepals are 13.2 mm long. The ovary is 7.33 mm long and 4.92 mm wide. The receptacle is 8.47 mm long and petal is 1.31 mm thick.

H. rosa-sinensis 'Tandang Sora' x *H. rosa-sinensis* 'Connie S. Angeles'

Fig. 1b shows the hybrid between *H. rosa-sinensis* 'Tandang Sora' and *H. rosa-sinensis* 'Connie S. Angeles'. It is a shrubby plant with a semi-erect growth habit. It is medium height at 1.29 m tall and 0.86 m wide. The leaves are ovate, with crenate margins, reniform base, apiculate apices and alternate petiole arrangement. The petiole is 18.4 mm long. The upperside of the leaves is glossy green (RHCC 137A), while the underside is green (RHCC 137B). It measures 85.9 mm long and 61.3 mm wide.

The flowers are solitary, funnel-shaped and borne in a slender stalk. The flower has involucral bracts. The bloom type is regular and simple with a bloom size of 128.7 mm. Flowers last for a day only, both as intact and cut flower, but succeeding blooms open each day. Corolla is 141 mm long, 123.8 mm wide, while the calyx lobe is 12.91 mm long and 10.56 mm wide. The floral angle of display on the plant is 83.64°. The corolla is tuscan yellow (RHCC 162C) with a reddish white (RHCC N1555A) halo and a ruby red (RHCC 59A) eye. The calyx is obovate, thick and imbricate. It is glabrous and light green (RHCC 141B). The pedicel is 45.4 mm long and the staminal tube is 65.6 mm. Five capitate stigmas that are orange (RHCC N25 A) are 2.02 mm long and 1.2 mm wide. The sepals are green and 11.1 mm long. The androecial type is monadelphous. The ovary is 8.09 mm long and 4.91 mm wide. The receptacle is 10.01 mm long. The petal is 1.42 mm thick. This hybrid has a tuscan yellow petals with ruby red eye.

H. rosa-sinensis 'Tarantella' x *H. rosa-sinensis* 'Golden Doubloom'

Fig. 1c shows the hybrid of 2 foreign varieties namely *H. rosa-sinensis* 'Tarantella' x *H. rosa-sinensis* 'Golden Doubloom'. The plant has a semi-erect growth habit, with medium height of 1.29 m and width of 0.68 m.

The leaves are ovate, has crenate margin, reniform base, apiculate apices and has an alternate petiole arrangement. The petiole is green and 33.6 mm long. The upperside of the leaves is glossy dull green (RHCC N137C), while the underside is light green (RHCC 137B). It is 121.3 mm long and 89.9 mm wide.

Flowers are solitary, funnel-shaped with involucral bracts and borne in slender stalks. The bloom type is simple and regular with a diameter of 138.6 mm. The flowers last for one day only, both as intact and cut flower, but succeeding bloom opens each day. The corolla is 14.9 mm long and 137.2 mm wide. It is red purple (RHCC 61B) with a yellow orange (RHCC 20A) edging and rose red (RHCC 58B) eye. The floral angle of display on the plant is 87.7°.

The calyx is obovate, thick, imbricate and green (RHCC 137B). The calyx lobe is 12.7 mm long and 11.26 mm wide. The pedicel is 36 mm long. The staminal tube is slender and 67.9 mm long. Five capitate stigmas are borne upon radiating branches. Stigmatic lobes are turkey red (RHCC 46C), 19.8 mm long and 11.9 mm wide. The sepal is green (RHCC 137B) and 1.65 mm long. The ovary is 10.29 mm long and 5.67 mm wide. The androecial type is monadelphous. The receptacle is 9.75 mm long, while the petal is 1.42 mm thick.

H. rosa-sinensis 'Perla Santos-Ocampo' x *H. rosa-sinensis* 'Loren B. Legarda'

Fig. 1d shows the hybrid between 2 Philippine-bred varieties, *H. rosa-sinensis* 'Perla Santos- Ocampo' and *H. rosa-sinensis* 'Loren B. Legarda'. It is shrubby with an erect growth habit. It grows to 2.09 m tall and 1.17 m wide. It is a fast grower on its own root.

The leaves are ovate, has crenate margin, attenuate base, acute apices and has an alternate petiole arrangement. The petiole is yellow green (RHCC 146C) and 36.2 mm long. The upper side of the leaves is green (RHCC N137B), while the underside is light green (RHCC N137D). The leaf is 107.7 mm long and 77.7 mm wide.

The flowers are solitary, funnel-shaped and borne in a slender stalk. The flowers have involucral bracts. The bloom type is regular and simple with a bloom size of 125.3 mm. Flowers have one-day longevity both as intact and cut flower, but succeeding flower buds open each day. The corolla is spanish orange (RHCC 26B), with a lemon yellow (RHCC 13B) eye and a dark orange (RHCC N 25B) vein markings radiating from the eye going to the petals. The corolla is thick, imbricate, obovate, 133.1 mm long and 120.3 mm wide. The glabrous calyx is green (RHCC 143C). Calyx lobe is 14.6 mm long and 12.53 mm wide. The pedicel is 60 mm long. The floral angle of display on the plant is 86.4°. The staminal tube is slender and 59.8 mm long. The capitate stigma has 5 lobes that are borne upon radiating branches. The stigmatic lobes are turkey red (RHCC 46C), 1.43 mm long and 0.8 mm wide. The sepal is green (RHCC 137B) and 1.4 mm long. The ovary is prominent, 7.86 mm long and 5.23 mm wide. The androecial type is monadelphous. The receptacle is 11.41 mm long while the petal is 1.24 mm thick.

H. rosa-sinensis 'Loren B. Legarda' x H. rosa-sinensis 'Tarantella'

Fig. 1e shows *H. rosa-sinensis* 'Loren Legarda x Tarantella', a product of cross-pollination of a foreign variety, *i.e.*, *H. rosa-sinensis* 'Tarantella' and a Philippinebred variety *H. rosa-sinensis* 'Loren B. Legarda'. The hybrid stands 0.81 m tall and 0.57 m wide with an erect growth habit.

Leaves are ovate, with crenate margin, apiculate apices, reniform base and alternate petiole arrangement. The petiole is yellow green (RHCC 146B) and 33.6 mm long. The leaf is 117.3 mm long and 90.8 mm wide. The upperside of the leaves is green (RHCC N137C), while the underside is light green (RHCC 137B).

The flowers are solitary, funnel shaped and borne in a slender stalk. Flowers have involucral bracts. The bloom type is simple and regular with 128.8 mm diameter. Flowers last for a day only, both as intact and cut flower, but succeeding flower buds open each day. Individual corolla is 141.4 mm long and 129.4 mm wide. The calyx lobe is 15.7 mm long and 10.75 mm wide. The angle of display of flower on the plant is 89.5°. The corolla color is a combination of primrose yellow (RHCC4B) and neyron rose (RHCC 58C).

The calyx is obovate, thick and imbricate. It is green (RHCC 143 A) and glabrous. The pedicel is green (RHCC 139D) and 31.6 mm long. The sepals are also green and 1.46 mm long.

The staminal tube is slender and measures 52.3 mm long. Five capitate stigma that are marigold orange (RHCC 28B) are borne upon radiating branches. The stigmatic lobes are 2.05 mm long and 1.14 mm wide. The androecial type is monadelphous. The ovary is 8.39 mm long and 5.59 mm wide. The receptacle is 10.28 mm long while the petal is 1.45 mm thick. Table 1. shows the summary of phenotypic characteristics of the 5 selected *H. rosa-sinensis* hybrids. The pedigree of the 5 *Hibiscus* hybrids were also indicated.

Correlation of floral and leaf characters of the Hibiscus hybrids

Correlation analysis of the different floral and leaf characters of the 5 selected hybrids indicated very high positive correlation on corolla length and bloom length (r = 0.94) (Table 2). High positive correlation was detected for corolla width and bloom length (r= 0.87), corolla width and corolla length (r= 0.87), style length and bloom length (r=0.71) and receptacle length and calyx lobe (r= 0.79). However, a negative high correlation was observed on sepal length and style length (r=-0.83).

A moderate positive correlation was detected for ovary width and bloom length (r = 0.5240), ovary width and corolla length (r = 0.5207), ovary width and corolla width (r = 0.5016) and style length and bloom length (r = 0.6579). In addition, moderate positive correlation was observed on the following: petiole length and receptacle diameter (r = 0.5430), petiole length and leaf width (0.6148) and leaf length and leaf width (r = 0.6486).

Pollen fertility

H. rosa-sinensis hybrids were subjected to pollen fertility testing by the staining method using potassium iodide (I_2 KI). Fully stained pollen is an indication of high viability (Fig. 2). Table 3 shows the pollen viability of the different hybrids with 61.62 to 70.0%. No significant differences were observed among the pollen viability of the different hybrids.

H. rosa-sinensis 'Accession 20' x *H. rosa-sinensis* 'Gelia Castillo' was subjected to pollen germination test using BK (1963) germination medium supplemented with varying concentration of sucrose as additive. The different sucrose concentrations added on the BK medium and the percent germination of *H. rosa-sinensis* 'Accession 20' x *H. rosa-sinensis* 'Gelia Castillo' were presented in Fig. 3. Germination was observed after incubation for 4 hours. Ten percent germination (60%) which is significantly different ($p \le 0.05$) to the percent germination at 20 and 10% sucrose. The lowest percent pollen germination was observed in BK medium with no sucrose.

Graft compatibility of different Hibiscus hybrids with two rootstock varieties

Fig. 4 shows the percent success of graft compatibility of selected H. rosa-sinensis hybrids when grafted to 2 rootstock varieties namely 'Reddy or Not' and 'Wilcox'. Significant difference ($p \le 0.05$) was detected between 2 varieties when used as rootstock for the hybrid H. rosa-sinensis 'Accession 20' x H. rosa-sinensis 'Gelia Castillo'. However, no significant difference was detected between the 2 rootstock varieties when the other hybrids were grafted onto them. Hybrid H. rosa-sinensis 'Accession 20' x H. rosa-sinensis 'Gelia Castillo' had 65% success of graft compatibility when grafted onto H. rosa-sinensis 'Wilcox' rootstock. However, 33% graft compatibility success was obtained when the same hybrid was grafted onto H. rosa-sinensis 'Reddy or Not'. However, H. rosa-sinensis 'Reddy or Not' and H. rosasinensis 'Wilcox' can still be used as rootstock variety for the other hybrids.

Discussion

Hybridization of Philippine native Hibiscus germplasm with the foreign or imported varieties aimed to improve the flower color and quality of the local varieties. This was done to upgrade the status of Philippine Hibiscus and rival it with other flowers in the market as a garden plant, potted ornamental and as landscaping material for subdivisions, parks, schools, golf courses, malls and other establishments. Collectively known as the University of the Philippines Los Baños Hibiscus Hybrids, the 44 hybrid varieties that were released from 1994 to 2016 were named after outstanding Filipino women achievers (Magdalita et al., 2009; Magdalita and Pimentel, 2010; Magdalita et al., 2011). Since the Hibiscus ornamental industry is very fashionable, constantly changing and further clamoring for new varieties, it is necessary to develop new hybrids with different colors and forms. Hence, this present research was conducted to characterize a new set of varieties that will be named after outstanding Filipino women in the field of education. This new series of Hibiscus hybrids will be called, "Women in Education Series" to commemorate the birth of the K to 12 education curriculum in the Philippines. The present study revealed a set of characteristics of the flower, leaf and the gross morphology of the hybrid plant. A similar set of characteristics was developed for the Hibiscus hybrids falling under the series 'Women in Public Service' and 'Women in Science' and 'Women Saints and Institutions Named after Them' (Magdalita and Pimentel, 2013; Magdalita et al., 2016). In Nigeria, a similar phenotypic characterization strategy was conducted for the edible Hibiscus sabdariffa or roselle, where the different accessions were characterized for their calyx color to be either red, deep red, light red or pink (Daudu et al., 2015).

The results of the characterization of the 5 selected Hibiscus hybrids indicated that each of them has a set of distinct characteristics that makes them easily distinguishable from each other from the existing hybrids reported in literature. One of the hybrids Hibiscus rosa-sinensis 'Accession 20' x H. rosa-sinensis 'Gelia Castillo' had solferino purple (RHCC 65B) petals with ruby red (RHCC 61A) eye, while the plant has medium height (1.37 m), erect growth with ovate and green leaves. This hybrid has similar purple or mauve flower color to H. rosa-sinensis 'St. Bridget College' (Magdalita et al., 2016), H. rosa-sinensis Kristie A. Kenny' (Magdalita et al., 2011), H. rosa-sinensis 'Star for All Seasons' (Magdalita and Pimentel, 2010) and a locallybred variety H. rosa-sinensis 'Cely Hermosa' (Magdalita et al., 2016). In comparison with the foreign-bred varieties, its flower color is also similar to H. rosa-sinensis 'Tahitian Taui', 'Tahitian Princess', 'Wagon Wheel', and 'Merv's Fascination' (Australian Hibiscus Society Inc., 2007, International Hibiscus Society Inc. nd). This hybrid inherited the purple petal color from the female parent H. rosa-sinensis 'Accession 20' while the red eye was inherited from the female parent H. rosa-sinensis 'Gelia Castillo'. This mode of inheritance of the red eye may suggest that red is dominant to pink. Similarly in salvia flowers (Salvia splendens), the red gene is dominant over pink or rose petal color (Robertson and Ewart, 1990).

Result of this study also showed that the hybrid between H. rosa-sinensis 'Tandang Sora' and H. rosa-sinensis 'Connie S. Angeles' had tuscan yellow (RHCC 162C) petals with ruby red (RHCC 59A) eye surrounded by reddish white (RHCC N1555A) halo. Its petal color is similar to the Philippine local hybrids H. rosa-sinensis 'Emerita V. De Guzman' (Magdalita and Pimentel, 2010) and has some resemblance to H. rosasinensis 'Estrella F. Alabastro' (Magdalita et al., 2009). Its petal color is very similar to the foreign variety H. rosasinensis 'Gwen Fraser' (Australian Hibiscus Society Inc., 2007). The tuscan yellow or white petal color was inherited from the female parent, indicating maternal inheritance, while the bright red eye is seemingly a combination of the different shade of red colors that were inherited from both parents. The result also indicated that the hybrid H. rosasinensis 'Tarantella' x H. rosa-sinensis 'Golden Doubloom' had red purple (RHCC 61B) petals with a yellow orange (RHCC 20A) edging and dark red rose (RHCC 58B) eye. Its petal color is unique and not resembling any of the parents indicating hybrid vigor. Hybrid vigor is a common phenomenon in many hybrids of plants having highly diverse parents (Brown and Caligari, 2008) like the parents of this hybrid, i.e., H. rosa-sinensis 'Tarantella' and 'Doubloom'. The hybrid's semi-spiral flower form was inherited from the female parent, indicating maternal inheritance of this trait. In addition, the dark red rose eye was inherited from the paternal parent H. rosa-sinensis 'Golden Doubloom'. This also suggests that red eye color is dominant to pink eye. Similarly in apple (Malus domestica) breeding, red peel color of a certain parent is dominant to other peel color, hence it is preferred for hybridization (Science Learning Hub, 2011; Janick and Moore, 1996). Furthermore, the present result jibed with the findings in tomato cv. Sta Clara, indicating that red peel is dominant to yellow (Do Rêgo et al., 1999).

Result of this study also revealed that the hybrid *H. rosa-sinensis* 'Perla Santos-Ocampo' x *H. rosa-sinensis* 'Loren Legarda' had spanish orange (RHCC 26B) petals with a lemon yellow (RHCC 13B) eye. This Spanish orange hybrid which is brighter orange in color resulted from a cross

between a light orange female parent (*i.e.*, *H. rosa-sinensis* 'Perla Santos-Ocampo') and a bright orange male parent (*i.e.*, *H. rosa-sinensis* 'Loren Legarda'). This suggests that the orange color in the female parent could have been intensified by the bright orange color coming from the male parent, suggesting additive effects. Petal color of this hybrid is similar to the introduced variety from Bangkok, Thailand *H. rosa-sinensis* 'Tangerine Orange' (Magdalita and Pimentel, 2013) and to the Philippine local hybrid *H. rosa-sinensis* 'Arlene B. Arcillas' x *H. rosa-sinensis* 'Marilyn D. Marañon' (Cabarrubias, 2015).

Furthermore, the result indicated that the hybrid between rosa-sinensis 'Loren Legarda' x H. rosa-sinensis H. 'Tarantella' had flower color combination of primrose (RHCC 4B) and neyron rose ((RHCC 58C). This new hybrid has a unique petal color combination that appeared to come from both parents, which could a form of hybrid vigor or heterosis. Heterosis is a common phenomenon that occurs in progenies of parents which are highly diverse (McMullen, 2008) like in F₁ intraspecific crosses of maize (Zea mays L.), radish (Raphanus sativus L.), cabbage (Brassica oleracea L.) and tomato (Lycopersicon esculentum L.) (Birchler et al., 2010). The primrose or dark pink color of the petals in this hybrid was inherited from the male parent, suggesting that pink could be dominant to yellow and orange. Similarly, in sweet cherry, (Prunus avium L.), it has been reported that red where the pink is included, is dominant to yellow (Janick and Moore, 1996). The semi-ruffled edges of the petals of the hybrid was also inherited from the male parent. Similarly, the Hibiscus hybrid progenies of H. rosa-sinensis 'Arlene B. Arcillas' x H. rosa-sinensis 'Marilyn D. Marañon' inherited the ruffled petal pattern of the male parent H. rosa-sinensis 'Marilyn D. Marañon' (Cabarrubias, 2015). In addition, H. rosa-sinensis 'Araceli A. Dans' and H. rosa-sinensis 'Che-Che Lazaro' inherited the ruffled petal edges from their parents whose petal edges are also ruffled (Magdalita et al., 2016). This may suggest that the ruffled petal edges trait is dominant over non-ruffled petal edges in Hibiscus.

Results of the correlation analysis of the different leaf and floral characters indicated a high positive correlation for corolla length and bloom length, corolla width and bloom length, corolla width and corolla length, style length and bloom length and receptacle length and calyx lobe. This suggests a strong association between these characters. In thalictrum (Thalictrum pubescens), another ornamental plant, correlation of floral traits was also conducted to trace evolution of the crop, wherein a high positive correlation was also detected for stigma length and ovary size (Davis, 2001). In contrast, result of the present study also indicated a negative high correlation between sepal length and style length among the Hibiscus hybrids evaluated. This suggests an inverse relationship between these 2 traits *i.e.*, a flower with short sepal will have a long style and vice-versa. This result jibed with the result of correlation of physico-chemical properties in mango (Mangifera indica L.) cultivars, wherein there was high negative correlation of fruit acidity with nonreducing sugars, total soluble solids (TSS), fruit weight, pulp content and sugar content (Bhowmick and Banik, 2008), which also implies that a fruit with low amount of acids will be sweet.

Correlation among phenotypic traits such as those with high and very high correlation suggests that a certain trait is highly associated to the other trait. Magdalita and Valencia (2004) used correlation analysis of selected fruit traits and found that fruit weight was correlated with fruit width. If a trait is correlated to another trait, only one could be evaluated, thus, this may lead to reduction of the number of characters to be evaluated during characterization of a new variety. If the data for the other associated traits are needed, extrapolation could be done to generate them if necessary through regression analysis. This reduction in the number of traits evaluated may allow researchers to evaluate more hybrids per unit time. Furthermore, in the Philippines, most of the *Hibiscus* hybrids are one-day old and considering the large number of hybrids for evaluation, considerable time and effort are needed. However, with the knowledge of correlated characters, this will lessen the number of traits to be evaluated; thus, it will simplify the evaluation process. In the present study, this basic information on correlated traits will be valuable during evaluation of numerous *Hibiscus* hybrids in the breeding programme.

For breeding and selection purposes for *Hibiscus* varieties with longer floral retention on the plant, correlation analysis of traits may provide essential information on the choice of parents to generate hybrids such as those with longer floral retention and bigger flower sizes. For instance, Valdoz (2015) found that peduncle diameter is very strongly correlated with length of floral retention. In the present study, the result revealed that receptacle diameter is highly correlated with the width of calyx lobe, which may suggest that flowers with wider calyx lobe will have longer floral retention.

Testing of pollen fertility in Hibiscus hybrid plants is important in determining the ability of the plant to fertilize another plant and produce viable seeds or fertilize itself, hence seeds are produced. An earlier report indicated that H. rosa-sinensis var. sinensis had good pollen fertility which is attributed to the polyploid nature of the plant (Cariño, 1992). Since hybrid progenies also become valuable sources of genetic materials for further hybridization, viable pollen should be present in the candidate parent materials. The present study indicated that the 5 new Hibiscus hybrids have fairly high pollen viability of 61.62 to 70.0% based on structural staining using I₂KI, but not significantly different. Based from this staining process, the highly stained pollen are considered viable, while the slightly stained and unstained ones are non-viable. This may suggests that the viable pollens contains carbohydrates, proteins and other molecules making them in good condition. Similarly, highly stained pollen of Leymus grass (Leymus chinensis) using I2KI were darkly stained with viability of 85.6% (Huang et al., 2004). Also, highly stained pollen of Tunisian caprifig var. Assafri (Ficus carica L.) were darkly stained with tetrazolium and had a viability of 84.0% (Gaaliche et al., 2013).

The result also showed that one of the hybrids H. rosasinensis 'Accession 20' x H. rosa-sinensis 'Gelia Castillo' exhibited good pollen germination of 60% on the BK medium added with 10% sucrose which is significantly different with the other sucrose concentrations used. This suggests that 10% sucrose added to the BK medium is optimum for pollen germination of *H. rosa-sinensis* 'Accession 20' x *H. rosa-sinensis* 'Gelia Castillo'. It has been reported that BK medium added with 10% sucrose is effective for pollen germination of many plant species (Brewbaker and Kwack, 1963). The present result corroborated with the previous report on Cleome (Cleome gynandra; Soni et al., 2010), Datura (Datura metel; Katara, 2002), Jathropa (Jathropa curcas) and Pedilanthus (Pedilanthus tithymaloides; Tiwari et al., 2014), indicating high pollen germination also using 10% sucrose. The result of the present study implies that the Hibiscus hybrids have fertile pollen capable of fertilization, hence they could be used as parents for further hybridization with other varieties.

Good pollen fertility is a positive characteristic of any plant that is contributory to the evalution of another plant. In the present study, germination of the pollen started 8 hours after incubation on the BK medium. This was manifested by the emergence of the pollen tube followed by elongation. Similar observation was reported in okra (*Hibiscus esculenta* L.) using artificial medium but supplemented with higher sucrose concentration of 20% instead of 10% (Baloch et al., 2001).

Testing of the graft compatibility of the 5 *Hibiscus* hybrids was conducted to evaluate their ability to unite with the 2 rootstock varieties namely *H. rosa-sinensis* 'Reddy or Not' and '*H. rosa-sinensis* Wilcox'. Propagation of the new hybrids will be made efficient with the use of a compatible rootstock coupled with the skills of the propagator and the cool season of the year. The result indicated significant difference ($p \le 0.05$) between *H. rosa-sinensis* 'Reddy or Not' and *H. rosa-sinensis* 'Wilcox' when grafted with the hybrid *H. rosa-sinensis* 'Accession 20' x *H. rosa-sinensis* 'Gelia Castillo'.

However, the result also indicated that there is no significant difference between the 2 rootstock varieties when grafted with the other hybrids. This suggests that H. rosasinensis 'Accesion 20' x H. rosa-sinensis 'Gelia Castillo' need to be grafted to H. rosa-sinensis 'Wilcox', but the other hybrids could be grafted to either H. rosa-sinensis 'Reddy or Not' or H. rosa-sinensis 'Wilcox'. This result jibed with the previous report of Magdalita and Pimentel (2013) who found that H. rosa-sinensis 'Wilcox' is the most suitable rootstock variety for grafting H. rosa-sinensis 'Cynthia Villar'. Further, this also conformed with the report of Little (n.d) who indicated that H. rosa-sinensis 'Ruth Wilcox' rootstock variety allows production of sturdy, robust and strong grafted plants which are fast growing when provided with ample amount of fertilizer. In addition, the present study used the modified side grafting technique, wherein the scion was inserted in between the bark and the wood of the rootstock where the cambial region is located. This technique allows the use of bigger rootstock about 1-2.1 cm diameter for small scion, and those scions of similar size to the rootstock, hence a greater flexibility in grafting and allowing efficient production of grafted plants. Similar technique was used successfully in grafting avocado (Persea americana Mill.; Adjei et al., 2005), and in the mass propagation of *H. sinensis* 'Cynthia A. Villar' (Magdalita and Pimentel, 2013).

Materials and Methods

Plant materials

The study was conducted in the *Hibiscus* Breeding Block at the Institute of Plant Breeding, College of Agriculture and Food Science, University of the Philippines Los Baños (14°09'13.5" N, latitude; 121°15'42.52" E, longitude), College, Laguna, Philippines from May 2014 to March 2015. Three-year old established *Hibiscus* hybrids generated from crosses between Philippine and foreign varieties were selected from the hybrids being maintained in the breeding blocks.

Hybridization

Hibiscus hybrids obtained from foreign sources such as Australia, Hawaii and Thailand such as like *H. rosa-sinensis* 'Tarantella' and *H. rosa-sinensis* 'Golden Doubloom' were crossed to local varieties such as *H. rosa-sinensis* 'Tandang Sora', 'Connie S. Angeles', 'Gelia Castillo', 'Loren B. Legarda', and 'Perla Santos-Ocampo'.

Designated female parent was identified and petals were removed to expose the stigma. The stigma was then covered with aluminum foil. On the succeeding sunny day, cross pollination was done. Successful fertilization allowed the enlargement of the ovary 2 weeks after cross pollination. The cross pollinated fruit was allowed to mature for a month and the seeds were then harvested and sown in a potting medium containing 1:1 (v/v) mixture of soil and sand. The germinated hybrid seedlings were allowed to grow for a month and they were transferred to individual pots. After 6 months, hybrid plants were transplanted to the breeding blocks. Selection of promising hybrids was done at the time of flowering when the hybrid plants are already one-year old. Unique flower color, with different color combinations and form, striking and bright eye surrounded by colorful halo and thick petals were some of the important criteria being considered in selection.

Phenotypic characterization

The plant stature, leaves and flowers were characterized qualitatively and quantitatively. The quantitative traits measured were plant height, bloom size, leaf length, leaf width, petiole length, pedicel length and angle of flower display on the plant. The qualitative traits evaluated included leaf color, flower color and leaf shape (Table 4).

The color of flowers and leaves were described based on the Color Chart of the Royal Horticultural Society of London (RHS 1966), other qualitative characters such as leaf shape and flower type were described based on the book "Vascular Plant Systematics" (Radford et.al., 1976).

Quantitative characters such as length and width of the flowers and leaves were measured using Vernier caliper and ruler. Plant height was measured using a meter stick, while the angle of flower display on the plant was measured using a protractor. Ten flowers and 10 leaves were used as samples to characterize each hybrid.

Pollen fertility of hybrids

Pollen fertility test of the 5 Hibiscus hybrids was conducted using the I₂KI structural staining test and pollen germination test on Brewbaker and Kwack's (1963) medium. Five selected hybrids were used for the pollen fertility testing. Fifty pollen samples were used for every replicate. Pollens were collected from 8:00 am to 10:00 am during sunny days only. Pollen grains were placed in 3 microscope slides with few drops of I₂KI solution. After 10 minutes incubation, pollen grains were observed under 100X magnification, using a digital microscope (True Vision Microscopes, Inc., USA). Darkly stained pollen was an indication of high pollen fertility, while unstained pollen was an indication of nonfertile pollen (Wani et al., 2015). Fully stained, partially stained and unstained pollen grains were counted for each of the 3 replications and percent viability was computed using the formula: % fertility = (Fully / darkly stained Pollen / Total number of pollen samples) X 100

In vitro pollen germination was also tested on Brewbaker and Kwack (BK) (1963). Pollen grains from the *H. rosa-sinensis* 'Accession 20' x *H. rosa-sinensis* 'Gelia Castillo' were germinated on this medium added with varying concentrations of sucrose at 0, 10, 20 and 30%. Three drops of BK medium were dispensed in depression slides and solidified for 5 minutes. Pollen grains were brushed on the medium and were germinated for 4 hours. The slides

containing the pollen grains were placed in plastic trays lined with moist tissue paper, and the trays were covered with plastic to conserve moisture. The samples were viewed under a light compound microscope (True Vision Microscopes, Inc., USA), and the germinated pollen grains were visualized in a computer aided by ToupTek ToupView 3.7 computer software. Pollens were considered germinated when the pollen tube was well-developed and elongated. Germinated and non-germinated pollens were counted, and the percent germination was computed for all the treatments in the 3 replications. The formula used was:

% germination = (Germinated Pollen / Total number of pollen samples) X 100

Graft compatibility of different Hibiscus hybrids using two rootstock varieties

Two different native *H. rosa-sinensis* varieties were used as rootstocks namely *H. rosa-sinensis* 'Wilcox' and *H. rosasinensis* 'Reddy or Not'. Semi-hardwood cuttings of these varieties were rooted in a medium containing a mixture of garden soil and coir dust (1:1, v/v). Two-month old rooted cuttings of *H. rosa-sinensis* 'Wilcox' and *H. rosa-sinensis* 'Reddy or Not' were grafted with the different hybrids. After grafting, the graft assembly was enclosed with a plastic bag following the '*Kulob*' system (enclosed system) that preserves and provides moisture to the developing shoot. The number of successful grafts was counted and the percent success of grafting was computed using the following formula:

% Grafting success = (No. of successful grafts / Total number of grafts) x 100

Fig. 5 shows the mother plant of *H. rosa-sinensis* 'Reddy or Not' and *H. rosa-sinensis* 'Wilcox' and the successfully grafted plants to the 2 rootstock varieties. These successfully grafted plants are one-month old.

Statistical analysis

The descriptive statistics such as the mean for quantitative traits and mode for qualitative traits of the 5 Hibiscus hybrids were used to characterize the hybrids. Fifty hybrid plants, *i.e.*, 10 plants for each hybrid were characterized, and for each plant sample, 10 flowers were used with a total of 500 flowers. Correlation analysis of floral and leaf characteristic was used to find traits associated with each other using the Pearson's Product Moment Correlation. The experiments on pollen fertility testing by structural staining and germination tests on BK medium (1963) were conducted using Completely Randomized Design (CRD) with 3 replications. Fifty pollen samples were used for each replicate. A11 percentage and count data were transformed accordingly prior to analyses. Analysis of variance (ANOVA) using the F-test at 0.05 level of significance was used to test the differences between treatment means. Pairwise mean comparison using the least significant difference (LSD) at a 0.05 level of significance was employed to test the differences between treatment means. On the other hand, the Z-test was used for analysis of the percent success of graft compatibility between the hybrids and the 2 rootstock varieties tested. The data analyses for all experiments were performed using Microsoft Excel 2010 and Statistical Tool for Agricultural Research 1.1 (STAR) (IRRI, 2014).

Conclusion

The 5 selected Hibiscus hybrids were characterized phenotypically. The hybrid H. rosa-sinensis 'Accession 20' x H. rosa-sinensis 'Gelia Castillo' is a solferino purple flower with ruby red eye surrounded by a grayish violet halo, while the hybrid H. rosa-sinensis 'Perla Santos Ocampo' x H. rosasinensis 'Loren B. Legarda' is a spanish orange flower with a lemon yellow eye surrounded by dark orange halo radiating from the eye going to the petal. The hybrid H. rosa-sinensis 'Tandang Sora' x H. rosa-sinensis 'Connie S. Angeles' is a tuscan yellow flower with a ruby red eye surrounded by reddish white halo, while H. rosa-sinensis 'Loren B. Legarda' x H. rosa-sinensis 'Tarantella' has a unique flower with the combination of primrose yellow and neyron rose. Furthermore, H. rosa-sinensis 'Tarantella' x H. rosa-sinensis 'Golden Doubloom' is a rose red flower with dark red rose eye and yellow orange edging on the petals.

Bloom size, corolla length and petiole length correlated positively with style length. Corolla width and bloom length also correlated positively with ovary width. Stigma width is positively correlated with stigma length. In addition, there was a high to very high positive correlation for corolla width and bloom length, corolla length and bloom length, leaf width and leaf length including petiole length and leaf width. The BK (1963) medium added with 10% sucrose significantly promoted pollen germination H. rosa-sinensis 'Accession 20' x H. rosa-sinensis 'Gelia Castillo'. Significant difference ($p \le 0.05$) in the pollen viability of the 5 Hibiscus hybrids was not detected using I2KI structural staining test. H. rosa-sinensis rootstock variety 'Wilcox' promoted grafting success by 60% when side-grafted with the hybrid H. rosa-sinensis 'Accession 20' x H. rosa-sinensis 'Gelia Castillo' than the rootstock variety H. rosa-sinensis 'Reddy or Not'.

Acknowledgements

The authors would like to thank the Institute of Plant Breeding, College of Agriculture and Food Science, University of the Philippines Los Baños. The assistance provided by Mr. Marcelino T. Gregorio, Mr. Jessie V. Silverio, Mr. Arcangel P. Cueto, Ms. Besseluz DC. Abayon, Ms. Maria Fe H. Cayaban, Ms. Lourdes Ann O. Angeles and Mr. Jonard C. Valdoz were also acknowledged. The funding support previously provided to the *Hibiscus* Breeding Programme by the Office of the former Chancellor Luis Rey I. Velasco was also acknowledged.

References

- Adjei PY, Boateng PY, Okyere Y (2005) Studies of three grafting methods of propagating avocado (*Persea americana* Mill.). Ghana J Hortic. 4:52-59.
- Australian *Hibiscus* Society, Inc (2007) *Hibiscus* growing guide handbook. 4th edn. Australian *Hibiscus* Society, Brisbane, Queensland, Australia. 40p.
- Baloch MJ, Lakho AR, Bhuto H, Solangi MY (2001) Impact of sucrose concentrations on *in vitro* pollen germination of okra (*Hibiscus esculentus*). Pak J Biol Sci. 4(4):402-403.
- Birchler JA, Yao H, Chubalayandi S, Vaiman D, Veitia RA (2010) Heterosis. The Plant Cell. 22:2105-2112.
- Bhowmick N, Banik BC (2008) Genetic variability and correlation studies for fruit physico- chemical properties of some mango cultivars grown under new alluvial zone of west Bengal. Asian J Hortic. 3(2): 346-349.

- Brewbaker JL, Kwack BH (1963) The essential role of calcium ion in pollen germination and pollen tube growth. Am J Bot. 50:859-865.
- Brown J, Caligari PDS (2008) An introduction to plant breeding. Blackwell Publishing Ltd Oxford OX4 2DQ, UK. 209p.
- Cabarrubias EMN (2015) Morphological characterization, evaluation and selection of *Hibiscus* (*Hibiscus rosasinensis* L.) hybrids. Unpublished undergraduate thesis, University of the Philippines Los Baños, College, Laguna, Philippines.
- Cariño ODC (1992) Cytology of gumamela (*H. rosa-sinesis Linn. var. sinensis*). Unpublished undergraduate thesis, University of the Philippines Los Baños, College, Laguna, Philippines.
- Cinco M (2013) Gumamela blooms as an economic driver. Available at: http://newsinfo.inquirer.net/165317/%E2%80%98gumamela%E2%80%99-blooms-as-economic-driver>. [Quoted on 2017- 4-15]
- Daudu OAY, Falusi OA, Dangana MC, Abubakar A, Yahaya SA and Abejide DR (2015) Collection and evaluation of roselle (*Hibiscus sabdariffa* L.). Afr J Food Sci. 9(3):92-96.
- Davis SL (2001) Phenotypic and genetic correlations among floral traits in two species of *Thalictrum*. J Hered. 92(2):361-366.
- Divya MJ, Sowmia C, Dhanya KP, Joona K (2013) Screening of antioxidant, anticancer activity and phytochemicals in methanolic extract of *Hibiscus rosasinensis* leaf extract. Res J Pharm Biol Chem Sci. 12(2):1308.
- Do Rêgo ER, Finger FL, Casali VWD, Cardoso AA (1999) Inheritance of fruit color and pigment changes in a yellow tomato (*Lycopersicon esculentum* Mill.) mutant. Genet Mol Biol. 22(1):101-104.
- Gaaliche B, Majdoub A, Trad M, Mars M (2013). Assessment of pollen viability, germination and tube growth in eight Tunisian caprifig (*Ficus carica* L.) cultivars. Int sch res notices Agron. 4pp. Avaialble at: http://dx.doi.org/10.1155/2013/207434>. [Quoted on 2017-4-15]
- Howie J (1980). *Hibiscus* Queen of the Flowers. 1st Edition. Brisbane, Australia. 72p.
- Huang Z, Zhu J, Mu X, Lin J (2004) Pollen dispersion, pollen viability and pistil receptivity in *Leymus chinensis*. Ann Bot- London. 93:295-301.
- International *Hibiscus* Society (nd). *Hibiscus rosa-sinensis* cultivars-International *Hibiscus* Society. Available at: https://www.google.com.ph/search?q=International+Hibiscus+Society&coq=In

ternational+*Hibiscus*+Society&aqs=chrome..69i57j0l2.124 88j0j4&sourceid=chrome&ie=UTF-

8#q=+site:international*Hibiscus*society.org+International+ *Hibiscus*+Society>. [Quoted on 2017-4-10]

- International Rice Research Institute (2014) Statistical analysis for agricultural research (STAR) package. Los Banos Laguna, Philippines.
- Janick J, Moore JN (1996) Fruit breeding volume 1 tree and tropical fruits. John Wiley and Sons, Inc. New York, USA. 614p.
- Katara M (2002) *In vitro* pollen germination of *Datura metel*. Int J Sci Res. 2(12):51-52.
- Kumar A, Singh A (2012) Review on *Hibiscus rosa-sinensis*. Int J Res Pharm Biomed Sci. 3(3):534-538.
- Little AL (n.d.) Hardwood cutting of Hibiscus. Hibiscusworld.com. Available at: http://Hibiscusworld.com/graft/prop1.htm. [Quoted on 2017-3-6]

- Magdalita PM, Valencia LD (2004) Fruit variability and correlation analysis of some phenotypic characters in avocado, rambutan and sweetsop. Philipp Agric Sci. 87(4):463-467.
- Magdalita PM, Gonzales-Lee VRC, Pimentel RB (2009) *Hibiscus* Hybrids Oblation Series: A Tribute to Outstanding University of the Philippines Alumnae for the University Centennial Year. Philipp J Crop Sci. 34(1):113-118.
- Magdalita PM, Pimentel RB (2010) *Hibiscus* Hybrids and Philippines' Women Achievers. National Academy of Science and Technology, Bicutan, Taguig City, Metro Manila. 58p.
- Magdalita PM, Gonzales-Lee VRC, Pimentel RB (2011) Development and horticultural characteristics of *Hibiscus* hybrids 'Women in Public Service Series'. Philipp J Crop Sci. 32(2):56-62.
- Magdalita PM, Pimentel, RB (2013) Development of *Hibiscus* hybrids "Women in Public Service Series II" and propagation studies on *Hibiscus rosa- sinensis* 'Cynthia A. Villar'. Philipp Sci Lett. 6(1):39-56.
- Magdalita PM, Cayaban MFH, Gregorio MT, Silverio JV (2016) Development and characterization of nine new *Hibiscus* hybrids. Philipp J Crop Sci. 41(2):31-34.
- McMullen (2008) Lecture Notes on Genetics and Crop Improvement – PLSC 646. North Dakota State University, Fargo, North Dakota, USA.
- Mishra N, Tandon VL, Munjal A (2009) Evaluation of medicinal properties of *Hibiscus rosa-sinensis* in male swiss albino mice. Int J Pharm Clin Res. 1(3):106-111.
- Naranja LR (2007) Floriculture: Sunshine industry in the Philippines. BAR Res Dev Dig. 9(1):1.
- Pabuyon TKM (2001) Prospects for the ornamental industry in the new millennium. BAR Res Dev Dig. 3(4):1.
- Pancho JV, Gruèzo WS (2006) Vascular Flora of Mount Makiling and Vicinity (Luzon: Philippines), Part 2. National Academy of Science and Technology (NAST) Philippines, Department of Science and Technology, Bicutan, Taguig City and Institute of Biological Sciences, College of Arts and Sciences, University of the Philippines Los Baños, College, Laguna. 626p.

- Pekamwar SS, Kalyankar TU, Jadhaya AC (2013) *Hibiscus rosa-sinensis*: A review on ornamental plant. World J Pharm and Pharm Sci. 2(6):4719-4727.
- Pimentel RB (1999) New cultivars and germplasm *Hibiscus* (*Hibiscus rosa- sinensis*) Centennial Series. Philipp Agric Sci. 82:238-244.
- Purseglove JW (1987) Tropical Crops. Dicotyledons. Singapore: Longman Singapore Publishers Pte. Ltd. 719p.
- Radford AE, Dickson WC, Massey JR, Bell CR (1976) Vascular Plant Systematics. Harper and Row, New York, USA. 640p.
- Royal Horticultural Society of London (1966) RHS Colour Chart 5th edn. Unpublished Standard Reference for Colors. Flower Council of Holland. Vincent Square, London.
- Robertson E, Ewart L (1990) Flower inheritance in *Salvia splendens*. Acta Hortic 272:9.
- Rummel DJ (2005) Botanical Beauty Book Compendium of Cosmetic Uses. C and E Pub Inc., Quezon City, Philippines. 433p.
- Science Learning Hub (2011) Breeding a new apple cultivar. Available at: https://www.science learn.org.nz/resources/844-breeding-a-new-apple-cultivar>. [Quoted on 2017-4-11]
- Soni, JN, Patel, MN, Jha CV (2010) Effect of different concentration of sucrose during *in vitro* pollen germination and pollen tube growth in *Cleome gynandra L*. Life Sci. (Leaflet).
- Tiwari A, Kamble RB, Chaturvedi A (2014) Pollen biology of some members of the Euphorbiaceae. Asian J Plant Sci. 4(6):8-14
- Valdoz JC (2015) Morph-anatomical characters and ethylene production in *Hibiscus rosa-sinensis* L. in relation to twoday old floral retention. Unpublished undergraduate thesis, University of the Philippines Los Baños, College, Laguna, Philippines.
- Wani TA, Pandith SA, Rana S, Bhat WW, Dhar N, Razdan S, Sharma W, Chandra S, Kitchlu S, Latto SK (2015) Promiscuous breeding behaviour in relation to reproductive success in *Grewia asiatica* L. Flora-Morphol Distrib Funct Ecol Plant 211: 65-71.