Flowering biology and pollination requirement in red pitaya (*Hylocereus* spp.)

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Abstract

This study explored the flowering biology and effects of pollination methods ((hand cross-, hand self-, automatic self-, and open-pollination) on fruit set, and fruit characteristics in 4 red pitaya *(Hylocereus* spp.) genotypes: [‘Vietnam White’ (‘VN-White’), ‘Chaozhou 5’, ‘Orejona’, and ‘F_{11}’] in Pingtung, Taiwan. The fruit production season started from the beginning of May and ended at the beginning of September with 6-7 flowering cycles and 22-32 flowers/plant/year. The flowering duration took from 15-19 days and fruit duration spent around 30 -32 days. The relative location of anthers were lower than the stigma, except in ‘VN-White’. ‘VN-White’, fully self-compatible, obtained high fruit set rates (80.0 - 90.5%) in all pollination treatments and the maximum fruit weight (402.6 g) in hand self- and (403.4 g) in open-pollination. ‘Chaozhou 5’ was partially self-compatible while ‘Orejona’ and ‘F_{11}’ were completely self-incompatible. Hand cross-pollination significantly increased fruit set and fruit weight in ‘Chaozhou 5’, ‘Orejona’ and ‘F_{11}’. Total soluble solid (TSS) content was not influenced by pollination methods.

Key words：*Hylocereus* spp., flowering biology, self-compatibility, pollination requirement.

INTRODUCTION

Pitaya or dragon fuit (*Hylocereus* spp.) originated from the Americas (Barthlott and Hunt, 1993) and has a wide distribution in tropical and subtropical regions (Mizrahi and Nerd, 1999; Merten, 2003). It is increasingly gaining interest in many countries, including Taiwan, due to its high economic potential as an exotic fruit crop and its exceptional tolerance to extreme drought (Raveh et al., 1998; Mizrahi and Nerd, 1999; Nobel and De La Barrera, 2004). Fresh dragon fruit is good for human health, providing essential nutrients such as vitamins, minerals, complex carbohydrates, dietary fiber and antioxidants (Le Bellec et al., 2006). Agronomic practices are relatively easy and inexpensive, due to fewer pests and disease attacks (Mizrahi and Nerd, 1999; Zee *et al*., 2004; Le Bellec *et al*., 2006).

In the world, pitaya genotypes which are being grown on commercial scale belong to four species: H. *undatus*, H. *monacanthus* (syn. H. polyrhizus), H. *costaricensis* and *Selenicereus megalanthus* (syn. *H. megalanthus*), and their hybrids (Weiss *et al*., 1994; Lichtenzveig *et al*., 2000; Tel-Zur et al., 2004 and 2011; Ortiz *et al*., 2012). Regarding reproductive biology, most studies reported that pitaya has large, hermaphroditic nocturnal flowers. It belongs to the long day plant with natural fruit production during warmer months (Mizrahi and Nerd, 1999; Merten, 2003). In Israel, *H. polyrhizus* and *H. costaricensis* were indicated as self-incompatible, requiring cross-pollination to set fruit, whereas *H. undatus* and *S. megalanthus* were self-compatible (SC), setting fruit with self-pollination (Weiss et al., 1994; Nerd and Mizrahi, 1997). In contrast to the findings of Weiss et al. (1994), under the natural habitat in Mexico, *H. undatus* had the highest fruit set after both open and unmanipulated self-pollination (Valiente Banuet *et al*., 2007). However, there have been no comprehensive studies on the effects of pollination on the performance of *Hylocereus* spp. growing under other conditions.

In Taiwan, red pitayas (*Hylocereus spp*.) with white, red or purple pulp are widely cultivated. Because of their importance as exotic fruit crops, a collection of different genotypes of the genera Hylocereus has been planted at the Tropical Fruit Orchard, at National Pingtung University of Science and Technology (NPUST). The present work investigated flower morphology, flowering phenology, and effects of pollination methods on fruit set and weight, TSS content in four typical or promising pitaya genotypes in order to determine their pollination requirement and to propose agro-management that can improve the efficiency of pollination, fruit quality and yield.

MATERIAL AND METHODS

2.1. Plant material and experimental sites

Four genotypes of pitaya (*Hylocereus* spp.) were used: 'VN-White' (*H. undatus*), 'Orejona' (*H. polyrhizus*), 'Chaozhou 5' and 'F_{11}' (*Hylocereus* sp.). The description of the four genotypes of pitaya used is presented in Table 1. Each plant was approximately 10 years old. The plants of these genotypes were intercropped with each other at NPUST. The experiment was conducted from May to September, 2013. The minimum and maximum daily temperatures during the experiment were 20.3°C (night) and 37°C (day), respectively.

Table 1. Descriptions of the four pitaya genotypes used for the pollination study

Genotype	Species	Fruit characteristics	Origin
'VN -White'	*Hylocereus undatus*	Oblong fruit with light-red peel, large scales, and white pulp	Vietnam
'Chaozhou 5'	*Hylocereus sp.*	Oblong-round fruit with dark-red peel, large scales, and violet-red pulp	Taiwan
'Orejona'	*Hylocereus polyrhizus*	Oblong fruit with dark-red peel, large scales, and violet-red pulp	Central America
'F_{11}'	*Hylocereus sp.*	Round fruit with dark-red peel, large scales, and violet-red pulp	Taiwan

2.2. Treatment and parameter measurements

2.2.1. Flowering biology

- Flowering phenoloy

Three plants for each genotype were monitored to determine flowering season, total flowers/plant/year, and the numbers of flowering cycles/plant/year. Flowers were considered as the same cycle when they opened on the same day. Five random floral buds for each genotype were labeled to identify duration of flowering and fruiting.

- Flower morphology characteristics

Size of five random mature flower buds for each genotype were measured and the flower morphological descriptors were identified at their fully opening stage. The descriptors and method of measurements are presented in Table 2.

Table 2. The flower morphological descriptors used in four red pitaya genotypes

Descriptor	Method
Mature flower bud size(cm)	Measure just before flower opening: Bud length, pericarpel length and width.
Sepal pattern	None, edged or striped
Petal color	White, yellowish green, yellow, creamor red
Stigma lobe color	Creamor green
Number of stigma lobes	Average number ofstigma lobes in 5 random flowers
Relative location of anthers and stigma	The distance between athers and stigma in fully opening flowers

2.2.2. Effect of pollination method

There were four pollination treatments. For hand cross-pollination, the anthers of mature flower buds were removed and the flowers were bagged before 14:00. These flowers were then hand-pollinated between 21:00–24:00 h with a fresh mix of pollen from the other three genotypes and re-bagged. For hand self-pollination, mature flower buds were covered with bags before 14:00, and pollen from the same flower was applied to the stigma after the flowers opened between 21:00–24:00. To prevent open pollination, the selfed flowers were bagged, except during pollination until 3 days ago later. For automatic self-pollination, the flowers were covered with bags for 3 days ago to prevent cross-pollination. For open pollination, natural pollination was allowed to occur without any interference.

The experiment was arranged in a randomized complete block design with three replications for each of the four genotypes selected. In each genotype, 15–18 flowers from these three plants (5 - 6 flowers per plant) in a row was considered as one replication. All mature fruits obtained (depending on the fruit set of each treatment) were analysed for their weight and sweetness. The rate of fruit set (percentage) was calculated as: (Number of fruits / Number of flowers) × 100 %. Fruit weights were measured using an electronic balance (GF-6100, A&D Company, Tokyo, Japan). TSS content of fruit flesh was measured using a hand refractometer (model PAL-1, Atago, Tokyo, Japan) and the result was expressed as °Brix.

2.3. Statistical analysis

Differences among means in pollination treatments were ascertained with Duncan's multiple range test, using Statistical SAS 9.0 software (Staitical Analysis System, SAS Institute, Cary, N.C.).

RESULTS AND DISCUSSION

3.1. Flowering biology

3.1.1. Flowering phenology

Dragon fruit belonges to the long day plant with natural flowering and production during warmer months (Mizrahi and Nerd, 1999; Merten, 2003). Under the conditions of Southern Taiwan, most genotypes tested started flowering in late April or early May except 'VN-White', which started in mid-May

(Table 3).

Table 3. Flowering season, flower cycles and numbers, flower and fruit stages in four red pitaya genotypes

Genotype	Flowering season	Flowering cycles/ year	Flowers /plant/year	Flowering duration (day)	Fruiting duration (day)
'VN -White'	3 month	6	22	15	31
'Chaozhou 5'	4 month	7	29	18	30
'Orejona'	3 month	6	26	18	32
'F_{11}'	3 month	6	32	19	32

Flowering season generally lasted three months including three flowering cycles. Only the flowering season of 'Chaozhou 5' continued for four months and consisted of seven flowering cycles. The flower number produced per plant in the year investigated varied from 22 to 32 (Table 3). The flowering duration ranged from 15 to 19 days and the fruiting duration generally took one month for all of the genotypes used (Table 3).

3.1.2. Flower morphology

The eight flower morphology descriptors are described in Table 3. The four genotypes tested have white-colored petals and cream colored stigma lobes. Three out of four genotypes have edged-sepal pattern except 'VN-white' (Table 4). 'Orejona' had the maximum mature bud size of 34.1±1.7 cm long, 14.5 ±0.2 cm of pericarpel length and 3.2±0.3 cm of pericarpel width whereas these minimum values were measured in 'F_{11}' with 28.6±1.6, 12.1±0.9, and 2.9±0.1 cm, respectively. The lowest number of stigma lobes was 21.7±3.8 per flower in 'Orejona', as compared with 28.0 to 28.7±1.5 cm in others. The upper part of the anthers located 1.7 cm (below the stigma) in 'Orejona', 1.5 cm in 'F_{11}' and 1.1 cm in 'Chaozhou 5', whereas the anthers in 'VN-White' were at the same height as the stigma. Similar to what had been found by Weiss et al. (1994), the position of anthers in three genotypes examined was lower than the stigma, although the distance varied between 1.1and1.7 cm (Table 4). The only exception is 'VN-White'. The anthers of 'VN-White' were at the same height as the stigma (Table 4).

Table 4. Morphological flower characteristics in four red pitaya genotypes Effect of pollination methods

Genotype	Mature bud			Opening flower				
	Bud length (cm)	Pericarp-el length (cm)	Pericarp-el width (cm)	Sepal patter -n	Petal color	Stigm a lobe color	Stigma lobe number	Anthers below stigma (cm)
' VN -White'	28.6 ±1.7	13.4±2.0	3.2±0.1	none	white	cream	28.7±1.5	0.1±0.3
' Chaozhou 5'	29.7±3.6	13.3±1.2	3.2±0.3	edged	white	cream	28.2±1.6	1.1±0.5
' Orejona'	34.1±1.7	14.5±0.2	3.2±0.3	edged	white	cream	21.7±3.8	1.7±0.5
' F_{11}'	28.6±1.6	12.1±0.9	2.9±0.1	edged	white	cream	28.0±0.0	1.5±0.4

3.2.1. Fruit set

Fruit set resulting from different pollination treatments of each pitaya genotype is indicated (Table 5). 'VN-White' was fully self-compatible, obtained high percentages of fruit set (80.0–95.2%) in all pollination treatments and the fruit set rate was not affected by pollination method. 'Chaozhou 5' was partially self-compatible due to no fruit formation in automatic self-pollination and a lower fruit set rate (52.2%) in hand self-pollination than open- and hand cross-pollination with 71.1 and 95.8 %, respectively. In contrast, 'Orejona' and 'F_{11}' were completely self-incompatible, only setting fruit by hand cross-pollination or by open-pollination. While 'Orejona' exhibited higher fruit set by hand cross-pollination than that of open pollination, 'F_{11}' set fruit equally well by both pollination treatments. When hand cross-pollination and open pollination was compared, hand cross-pollination always had higher fruit set regardless of genotype.

Table 5. Effect of pollination method on fruit set (%) in four pitaya genotype

Genotype	Hand cross-pollination[#]	Hand self-pollination	Auto. self-pollination	Open pollination
VN -White	85.6 a A[†]	95.2 a A	80 A	90.5 ab A
Chaozhou 5	95.8 a A	52.2 b C	–	71.1 bc B
Orejona	88.4 a A	–	–	61.1 c B
F_{11}	90.2 a A	–	–	91.7 a A

[#] Crossing with mixed pollen from the other three genotypes.
[†] Values are the means of three replications per treatment (n = 45–54). Mean values followed by the same lower-case letter in each column, or with the same upper-case letter within each row were not significantly different at $P \leq 0.05$ by Duncan's multiple range test.
–, no fruit set.

Weiss et al. (1994) concluded that self-incompatible species had high fruit set after being cross-pollinated. This finding was also true for two of our genotypes. However, Weiss et al. (1994) reported that self-compatible species (*H. undatus*) had lower fruit set percentage after open- pollination (43%) than hand cross-pollination (100%) and no fruit set in automatic self-pollination. This is contradictory to what we found. These differences may be due to the anthers and stigma being close in VN-White and at least 2 cm apart in the H. undatus clones they used. Because their H. undatus clones did set fruits using hand self-pollination, (50-79.6%), it appears that pollen vectors are necessary for their clones. VN-White is completely autogamous, possibly due to its long selection in Vietnam.

3.2.2. Fruit weight

The fruit weight was differently affected by pollination treatments among genotypes (Table 6). In fully self-compatible genotype 'VN-White', the heaviest fruits were obtained from hand self- and open-pollinations with 402.6 and 403.4 g, respectively, whereas automatic self-pollination produced the lightest fruit and hand cross-pollination resulted in the intermediate values. In contrast, in the other three partially–completely self-incompatible genotypes, hand cross-pollination produced significantly larger fruits than open-pollination, which ranged from 281.8–416.3 g, compared to 145.0–295.8 g, respectively.

Table 6. Effect of pollination method on fruit weight (g) in four red pitaya genotypes

Genotype	Hand cross-pollination	Hand self-pollination	Auto. self-pollination	Open-pollination
VN -White	326.1ab[†]	402.6 a	275.1 b	403.4 a
Chaozhou 5	374.2 a	251.4 b	–	295.8 b
Orejona	281.8 a	–	–	149.2 b
F_{11}	416.3 a	–	–	145.0 b

[†] Values are the means of three replications per treatment (n = 24–49, all mature fruits obtained from the same treatments shown in Table 5 were measured). Mean values followed by the same lower-case letter in each row were not significantly different at $P \leq 0.05$ by Duncan's multiple range test.
- no fruit set.

The efficiency of pollination is reflected by fruit set and weight. Fruit weight in cacti was correlated with the number of seeds (Weiss et al., 1994). Open-pollination gave poorer fruit set, compared with that of hand cross-pollination in 'Chaozhou 5' and 'Orejona', and smaller fruit in 'Chaozhou 5', 'Orejona' and 'F_{11}'. However, these two treatments did not affect fruiting in 'VN-White' (Table 5 and Table 6). One explanation for this difference could be the differences in the mating system related to the relative location of the anthers and stigma.

3.2.3. Total soluble solid content

There was no significant difference in TSS content between hand cross- and open-pollination in 'Orejona' (17.5 and 17.0°Brix) and 'F_{11}' (17.1 and 17.7 °Brix), respectively (Table 7). However, this parameter tended to have higher values in smaller fruits of other genotypes. In 'VN-White', auto self-pollinated fruits contained 19.4 °Brix, as compared with 18.2 °Brix in hand self- and open pollinated fruits whereas 'Chaozhou 5' fruits obtained 18.4 °Brix in hand self- and 18.7 °Brix in open-pollination, as opposed to 16.7 °Brix in hand cross-pollination.

Table 7. Effect of pollination method on TSS content (°Brix) in four red pitaya genotypes

Genotype	Hand cross-pollination	Hand self-pollination	Auto. self-pollination	Open-pollination
VN -White	18.8 ab[†]	18.2 b	19.4 a	18.2 b
Chaozhou 5	16.7 b	18.4 a	-	18.7 a
Orejona	17.5	-	-	17.0
F_{11}	17.1	-	-	17.7

[†] Values are the means of three replications per treatment (n = 24–49, all mature fruits obtained from the same treatments shown in Table 5 were measured). Mean values followed by the same lower-case letter in each row were not significantly different at $P \leq 0.05$ by Duncan's multiple range test.

- no fruit set.

CONCLUSIONS

The natural fruit season of dragon fruit in Pingtung is concentrated in 3 summer months, from June to August with 6-7 flower cycles and 22–32 flowers/plant/year. It takes 46 to 51 days for flowering-fruit harvest duration. 'VN-White' was fully self-compatible and produced high fruit set and fruit weight by both open- and hand self pollination, which will lead to an advantage in single cultivar orchards without the need for manual supplementary pollination. Three red flesh genotypes: 'Chaozhou 5', 'Orejona' and 'F_{11}' are partially self- or completely incompatible, requiring cross pollination to set fruits. In cases of natural pollination inefficiency, hand cross pollination will ensure higher fruit set and heavier fruit weight in these three genotypes.

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紅龍果開花生物學及授粉需求之研究

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摘 要

本研究探討四個紅龍果基因型在屏東的開花生物學及授粉方法(人工雜交、自交、自然自交及開放授粉)對著果及果實性狀之影響。其開花期約在5月初至9月初之間，每年每株有6-7批花以及22-32朵花。花苞孕育期約15-19天，而果實生長期約30-32天。除了‘越南白肉’之外，其它品種花藥的相對位置均低於柱頭。完全自交親和之‘越南白肉’在所有授粉處理中均有高的著果率(80.0 – 90.5%)，且在人工自交及開放授粉下有最大果重。‘潮州5號’為部分自交親和，而‘Orejona’及‘F_{11}’為完全自交不親和。人工雜交授粉顯著增加‘潮州5號’、‘Orejona’及‘F_{11}’之著果率。授粉處理對果肉可溶性固形物並無顯著影響。

關鍵字：紅龍果(*Hylocereus* spp.)、開花生物學、自交親和性、授粉需求