Annals of Botany **93**: 469–472, 2004
doi:10.1093/aob/mch055, available online at www.aob.oupjournals.org

Metaxenia in the Vine Cacti *Hylocereus polyrhizus* and *Selenicereus* spp.

YOSEF MIZRAHI[1,2,*], JOSEPH MOUYAL[2], AVINOAM NERD[2] and YARON SITRIT[2]

[1]*Department of Life Sciences and* [2]*The Institute of Agriculture & Applied Biology, Ben-Gurion University of the Negev, PO Box 653, Beer-Sheva 84105, Israel*

Received: 7 July 2003 Returned for revision: 18 November 2003 Accepted: 10 December 2003

• *Background and Aims* Flowers of the vine cacti of the genera *Hylocereus* and *Selenicereus* grown in Israel must be hand pollinated due to self-incompatibility and lack of efficient pollinators. In controlled pollination experiments, it was found that the time elapsed between pollination and ripening depends on the source of the pollen. Therefore a study was made of some effects of the pollen source on fruit development.
• *Methods* Flowers of *Hylocereus polyrhizus* were pollinated on the same day with different pollen sources and the stigmas were covered. Fruits were collected 4 d after reaching full colour.
• *Key Results* Pollinating flowers of *Hylocereus polyrhizus* with *Selenicereus grandiflorus* and *S. megalanthus* pollen delayed ripening by 1 and 3 weeks, respectively, as compared with ripening of fruits pollinated with *Hylocereus undatus* pollen. Other fruit characteristics affected by the pollen source were fruit size, pulp dry weight and number of seeds per fruit, all being significantly reduced, while peel dry weight was significantly increased by *S. megalanthus* pollen. Total soluble sugar content was reduced in *H. polyrhizus* fruits pollinated with *S. grandiflorus* pollen. No other major traits were affected.
• *Conclusions* The results are evidence for the existence of metaxenia, i.e. an effect of pollen on maternal tissues, in cacti fruits. This pollen effect on the fruit-ripening time may be used for extending the marketing period of *H. polyrhizus* fruits.

Key words: *Hylocereus polyrhizus*, *Hylocereus undatus*, *Selenicereus megalanthus*, *Selenicereus grandiflorus*, metaxenia, xenia, pitaya, ripening, fruit, Cactaceae.

INTRODUCTION

Vine cacti, mainly of *Hylocereus* and *Selenicereus* genera, are newly developed fruit crops in Israel (Mizrahi and Nerd, 1999; Nerd *et al*., 2002). Due to the lack of efficient pollinators in Israel, manual cross-pollination is necessary to guarantee fruit set (Weiss *et al*., 1994; Mizrahi *et al*., 1997; Nerd and Mizrahi, 1997). Self-incompatibility among the *Hylocereus* and *Selenicereus* genera is common and therefore efficient fruit set is achieved only by cross-pollination with compatible pollen groups. While performing experiments to define cross-pollination compatible groups, we found that the source of pollen affected the time required for fruit development. This phenomenon of pollen affecting fruit characteristics has been known as xenia (Focke, 1881, as reviewed by Denney, 1992; Sedgley and Griffin, 1989). However, there is confusion in the usage of the terms xenia and metaxenia and it has been suggested that 'xenia' should be used to describe pollen effects on tissues that contain at least one set of genes from the male, namely the embryo and the endosperm, whilst the term 'metaxenia' should be used to describe the effects of pollen on fruit tissues of maternal origin, such as the pericarp and other fruit components that have no male contribution (Denney, 1992). Since fruit ripening in vine cacti is correlated with peel colour change from green to deep red (Nerd *et al*., 1999), it was obvious that we were dealing with metaxenia. While searching the scientific literature, we were surprised to learn that there are only a few papers dealing with metaxenia, and practically no data concerning cacti plants (Osman *et al*., 1974; Daulta and Chauhan, 1983, 1984; Vashishtha, 1986; Purohit, 1987; Al-Ghamdi *et al*., 1988; Chaudhary and Desai, 1995; Chen QH *et al*., 1996; Rahemi, 1998).

In the present paper, the occurrence of metaxenia is described in vine cacti fruits and it is suggested that this phenomenon may be used for extending the marketing period by controlling the ripening time.

* For correspondence. Fax 972–8-6472984, E-mail mizrahi@bgumail.bgu.ac.il

MATERIALS AND METHODS

Plant material

Flowering plants of *Hylocereus polyrhizus* (Weber) Britt. & Rose clone 89-028, *Selenicereus grandiflorus* (L.) Britt. & Rose clone 92-080, and *Selenicereus megalanthus* (Schum.) Britt. & Rose clone E2 were used as fruit producing plants and as sources of pollen (Mizrahi and Nerd, 1999). The plants were 5–6 years old and grown in our genotype-collection glasshouse. *Hylocereus undatus* (Haworth) Britt. & Rose clones compatible with *H. polyrhizus* were used as the pollen source for fertilizing the self-incompatible *H. polyrhizus*. All *Hylocereus* species used are fertile diploid (Lichtenzveig *et al*., 2000). It was observed in different pollination experiments that all *Hylocereus* species (*H. undatus*, *H. costaricensis* and *H. polyrhizus*) yielded similar results. Therefore, data combined from these experiments are presented using the term *Hylocereus* spp.

to describe the pollen source. Both *S. grandiflorus* and *S. megalanthus* are self-compatible.

Growing conditions

Plants were grown under 60 % shade, as previously described (Raveh *et al*., 1998). Since the flowers open at night and stay open until the following morning only (Weiss *et al*., 1994; Nerd and Mizrahi, 1997), pollen was collected early at night upon flower opening and applied with a paintbrush to the stigmas in the early morning. After pollination, the stigmata were immediately covered with cheesecloth bags to avoid unwanted pollination. Fruits were collected 3–4 d after reaching full colour and analysed for fruit components, number of seeds and chemical composition, as specified by Nerd *et al*. (1999).

Fruit analysis

Total acids were measured in aqueous pulp extracts. Five grams of pulp tissue were homogenized with 5 mL of double-distilled water with a Polytron (PCU, Lucerne, Switzerland). The shaft was washed with an additional 10 mL of double-distilled water, which was then added to the homogenates. After centrifugation (10 000 *g*, 10 min) the pH of the supernatant was determined and total acidity was measured by titration with 0·1 N NaOH to pH 8·2.

Total soluble solids were determined using a refractometer (PR-100, Atago, Japan) on diluted sap that had been expressed from the pulp.

Total soluble sugars concentration was determined in 50 mg samples of dry pulp by the phenol–sulphuric acid method as described previously (Nerd and Mizrahi, 1998).

Statistics

Data were analysed by ANOVA and Student's *t*-test (SAS Institute, Cary, USA).

RESULTS

The most obvious evidence of metaxenia in vine cacti is the pollen effect on the time required for fruit development, as seen in Fig. 1, which depicts two fruits of *H. polyrhizus* on the same branch at different stages of ripening. These fruits developed from flowers that had been pollinated on the same day with different sources of pollen: the red, ripe fruit originated by pollination with pollen of *H. undatus* clones, while the green, unripe fruit resulted from pollination with *S. grandiflorus* pollen. The ripening-time difference between the two fruits was about 5 d (Table 1). The data presented in Table 1 also show that the pollen source had an effect on fruit weight and this latter effect is also seen in Table 2. Pollination of *H. polyrhizus* and *S. grandiflorus* with pollen of *H. undatus* clones caused earlier ripening and smaller fruits than pollination with *S. grandiflorus* pollen (Table 1). *Hylocereus polyrhizus* fruit size was even more reduced when pollinated with pollen of the tetraploid plant *S. megalanthus*, and the period required for fruit development was extended by 3 weeks (Tables 1 and 2).

FIG. 1. *Hylocereus polyrhizus* branch bearing two fruits at different stages of ripening. The red, ripe fruit originated from pollination with pollen of *H. undatus* clones, while the green, unripe fruit was from pollination with *S. grandiflorus* pollen. The photograph was taken 34 days after pollination.

Among the fruit characteristics studied in *H. polyrhizus*, e.g. percentage of pulp and percentage of dry matter in the peel and pulp, the latter trait was most affected by the pollen source (Table 2). The influence of the pollen source on dry weight was most significant for *S. megalanthus* pollen, resulting in the lowest value in the pulp and the highest value in the peel. Values for the other two pollen sources, *H. undatus* clones and *S. grandiflorus*, were similar (Table 2). *Selenicereus megalanthus* pollen also significantly reduced the portion of pulp fresh weight, yielding fruit with thicker peel as compared with *H. undatus* pollen (Table 2). The number of seeds per fruit, as well as per gram of pulp, was also affected by the pollen source, being the smallest for *S. megalanthus* pollen; however, the effect on seed weight was not significant (Table 3).

No influence of the pollen source was found on ripening parameters affecting fruit quality, e.g. total soluble solids (TSS), acidity (Table 4) and taste (data not shown). However, in fruits developed by pollination with *S. grandiflorus* the level of total soluble sugars was significantly lower than fruits developed by pollination with *Hylocereus* spp. pollen (Table 4). These results imply that only certain fruit traits are affected by the pollen source, and the overall quality of the fruits was not affected.

DISCUSSION

The results show that the vine-cactus fruits exhibit metaxenia, since the tissues affected by the pollen source are solely of maternal origin (Osman *et al*., 1974; Daulta and Chauhan, 1983, 1984; Kahn *et al*., 1994; Chaudhary and Dessay, 1995; Nerd and Mizrahi, 1997).

The effect of the pollen source on the maternal tissue might be mediated by the seeds, which serve as a source for plant hormones. Swingle first raised this idea as long ago as 1928, when he tried to explain metaxenia in the date palm. If the seeds regulate the levels of hormones reaching the maternal tissues, they directly control events in the maternal

TABLE 1. *Effect of pollen source on fruit weight and days required for fruit ripening*

Female plant	Pollen source	*n*	Fruit weight (g)	Days to ripening
H. polyrhizus	*H.* spp. clones	66	370·4[b]*	31·4[c]
	S. grandiflorus clones	38	470·4[a]	36·1[b]
	S. megalanthus	4	224·3[c]	51·0[a]
S. grandiflorus	*H.* spp. clones	18	66·7[b]	54·5[b]
	S. grandiflorus	12	101·8[a]	60·2[a]

* Means within a column followed by different letters are significantly different at $P < 0{\cdot}05$.

TABLE 2. *Effect of pollen source on various characteristics of* Hylocereus polyrhizus *fruits*

Pollen source	*n*	Fruit weight (g)	Pulp (%)	Dry weight (%)	
				Peel	Pulp
H. undatus clones	8	391·8[a]*	58·7[a]	8·0[b]	17·0[a]
S. grandiflorus	8	461·2[a]	56·2[ab]	7·5[b]	16·3[a]
S. megalanthus	4	224·3[b]	53·2[b]	9·4[a]	13·9[b]

* Means within a column followed by different letters are significantly different at $P < 0{\cdot}05$.

TABLE 3. *Effect of pollen source on seed number and seed weight in fruits of* Hylocereus polyrhizus

Pollen source	*n*	No. of seeds/fruit	No. of seeds g^{-1}	Av. seed wt (mg)
H. undatus clones	8	4744[a]*	20·9[a]	2·41
S. grandiflorus	8	4362[a]	16·8[b]	2·57
S. megalanthus	4	735[b]	6·1[c]	2·71

* Means within a column followed by different letters are significantly different at $P < 0{\cdot}05$.

TABLE 4. *Effect of pollen source on total soluble solids, total soluble sugars and acidity in pulp of* Hylocereus polyrhizus *fruits*

Pollen source	*n*	TSS (%)	Sugar (mg g^{-1})	Acidity (µmol H^+ g^{-1})
H. spp.	8	13·1	84·2[a]*	492
S. grandiflorus	8	14·1	70·5[b]	499
S. megalanthus	4	13·9	79·7[ab]	491

* Means within a column followed by different letters are significantly different at $P < 0{\cdot}05$.

tissues of the developing fruit, including the time required for ripening (Denney, 1992). The effect of the pollen source on fruit development time has been reported for several species from different families, including date palm (Swingle, 1928), raspberry (Colber and de Oliveria, 1990), blueberries (Gupton and Spiers, 1994), cherry and pear [as cited from Stancevic (1971) and Nyeki (1972), respectively, by Gupton and Spiers (1994)] and cherimoya (Khan *et al*., 1994). Denney (1992) defined the effect of the pollen source on the timing of fruit ripening as 'xenochrons'. This term fits the phenomenon described above in *Hylocereus*. Where the time which elapses between pollination and ripening is affected by the pollen source (Table 1). Pollen of the two species of the genus *Selenicereus, S. grandiflorus* and *S. megalanthus,* prolonged the time to ripening of *H. polyrhizus* fruits. This effect may be due to the longer period required for fruit development in these two species compared with *H. polyrhizus* fruit (Lichtenzveig *et al*., 2000; Tel-Zur, 2003). Similar results were obtained in blueberries, in which fruit development time correlated with that of the clones used as the pollen source (Gupton and Spiers, 1994; Gupton *et al*., 1997). However, recently, Ehlenfeldt (2003) reinterpreted these results to suggest that in blueberry pollen source ability to enhance or delay ripening is the outcome of fertility problems. This was not the case in the vine cacti since viable seed numbers were similar in *H. polyrhizus* fruits developed from either *S. grandiflorus* pollen or *Hylocereus* spp. pollen (Tables 1 and 3).

The differences in the time to fruit ripening might be explained by variable growth rates of the pollen tubes. However, in previous work it was found that pollen tubes of *S. megalanthus*, *H. polyrhizus* and *H. undatus* completed germination and reached the ovary 48–96 h after pollination (Lichtenzveig *et al*., 2000). Therefore the difference of 20 d in fruit ripening (Table 1) cannot be attributed to a difference in growth rates of the pollen tube. Moreover, comparing pollen tube growth rates in conspecific crosses versus self-pollination in the self-compatible *S. megalanthus* and *H. undatus* revealed that germination and growth rates were similar (Lichtenzveig *et al*., 2000). The reduction of seed number in *H. polyrhizus* fruits produced from pollination with *S. megalanthus* pollen should not be considered xenia, because the latter is a semi-sterile tetraploid species having pollen widely ranging in size with low viability and germinability (Lichtenzveig *et al*., 2000) caused by unbalanced meiosis (Tel-Zur *et al*., 2003). Thus the considerable inviability of *S. megalanthus* pollen can be attributed as the cause of the reduction in seed numbers per fruit. Reduction of seed number in turn reduces the fruit size, which generally correlates with seed number (Weiss *et al*., 1994; Nerd and Mizrahi, 1997). The other *Selenicereus* species used in the experiments, *S. grandiflorus*, which is a diploid producing normal pollen, significantly reduced the number of seeds per unit weight of fruit, but nonetheless increased fruit weight. It is therefore worth trying to use pollen of other species of *Selenicereus* to avoid the reduction in fruit size while retaining the beneficial effect on fruit development time.

Other metaxenia effects that were found, e.g. on percentage of dry material in various tissues and sugar accumulation, may also be explained by seed-controlled hormonal level. This again highlights that the role of plant hormones in metaxenia should be investigated. Unfortunately little is known about hormone regulation of fruit development and ripening in vine cacti.

No other effects of metaxenia in its wider definition were found in the vine cacti studied. Fruit shape, colour, and taste were similar, regardless of the sources of pollen used in these experiments. However, the effect of pollen source on sugar accumulation should be carefully examined since the pollen source might affect the taste qualities of the fruit. These results are similar to those reported for dates, cherimoya, grapes and mandarins, in which the source of pollen had an effect on maternal tissue characteristics (Denney, 1992; Wallace and Lee, 1999).

Although the principal cause of metaxenia is not unambiguously understood, this phenomenon may be put to immediate use in horticultural practice. The vine cacti usually flower in waves, resulting in fluctuating quantities of ripe fruits from month to month, about 1 month apart. For a good marketable crop, a constant supply of fruits throughout the season is required (Mizrahi and Nerd, 1999). Due to their short shelf-life, storing these fruits is currently not practical (Nerd *et al*., 1999). One way to lengthen the marketing season could be to pollinate the fruits with pollen sources that affect the ripening time differently. The time delay in ripening is equivalent to extending the shelf-life by the same length of time. Such a technique is feasible in Israel, where all fruits are hand-pollinated (Mizrahi and Nerd, 1999; Nerd *et al*., 2002), but not where open pollination is the common practice. A similar idea whereby out-of-season (early) fruits could be produced through the use of different pollen sources has already been suggested for rabbiteye blueberries (Gupton and Spiers, 1994).

ACKNOWLEDGEMENTS

We thank Ms Dorot Imber for editing the manuscript. We also thank the Shirley and William Fleischer Foundation and the Arline and Ben Gueren Foundation for their partial support of this project. This work was partially supported by the Chief Scientist Ministry of Agriculture, Israel (grant no. 857-0414-02)

LITERATURE CITED

Al-Ghamdi AS, Al-Hassan GM, Jahjah M. 1988. Evaluation of eight seedling date palm (*Phoenix dactylifera* L.) males and their effects on fruit character of three female cultivars. *Arab Gulf Journal of Scientific Research, B Agricultural and Biological Sciences* **6**: 175–187.

Chaudhary SM, Desai UT. 1995. A short note on metaxenia in mango. *Recent Horticulture* **2**: 147–148.

Chen QH, Zhang ZH, Qin ZQ, Jiang YCh, Chen QH, Zhang ZH, Qin ZQ, Jiang YC. 1996. Study on the selection of male plants for Jinkui kiwifruit, and pollen xenia. *China Fruits* **2**: 23–24.

Colbert S, de Oliveira D. 1990. Influence of pollen variety on raspberry (*Rubus idaeus* L.) development. *Journal of Heredity* **81**: 434–437.

Daulta BS, Chauhan KS. 1983. Metaxenia studies on fruit set, maturity and quality characters in grapes (*Vitis vinifera* L.). *Haryana Journal of Horticultural Sciences* **12**: 22–25.

Daulta BS, Chauhan KS, 1984. Metaxenia studies on some berry and seed characters in grapes (*Vitis vinifera* L.). *Indian Journal of Horticulture* **41**: 73–79.

Denney JO. 1992. Xenia includes metaxenia. *HortScience* **27**: 722–728.

Ehlenfeldt MK. 2003. Investigations of metaxenia in northern highbush blueberry (*Vaccinium corymbosum* L.) cultivars. *Journal of the American Pomological Society* **57**: 26–31.

Focke WO. 1881. *Die Pflanzen Mischinge: ein Beitrag zur Biologie Der Gewächse*. Berlin: Borntraeger, 510–518 (cited by Denny, 1992).

Gupton CL, Spiers JM. 1994. Interspecific and intraspecific pollination effects in rabbiteye and southern highbush blueberry. *HortScience* **29**: 324–326.

Gupton CL, Yarborough DE; Smagula JM. 1997. Evidence of xenia in blueberry. Proceedings of the sixth international symposium on *Vaccinium* culture, Orono, Maine, USA. *Acta Horticulturae* **446**: 119–123.

Kahn TL, Adams CJ, Arpaia ML. 1994. Paternal and maternal effects on fruit and seed characteristics in cherimoya (*Annona cherimola* Mill.). *Scientia Horticulturaee* **59**: 11–25.

Lichtenzveig J, Abbo S, Nerd A, Tel-Zur N, Mizrahi Y. 2000. Cytology and mating system in the climbing cacti *Hylocereus* and *Selenicereus. American Journal of Botany* **87**: 1058–1065.

Mizrahi Y, Nerd A. 1999. Climbing and columnar cacti – new arid lands fruit crops. In: Janick J. ed. *Perspective in new crops and new crops uses*. Alexandria, VA: ASHS Press, 358–366.

Mizrahi Y, Nerd A, Nobel PS. 1997. Cacti as crops. *Horticultural Review* **18**: 291–320.

Nerd A, Mizrahi Y. 1997. Reproductive biology of cactus fruit crops. *Horticultural Review* **18**: 321–346.

Nerd A, Mizrahi Y. 1998. Fruit development and ripening in yellow pitaya. *Journal of the American Society for Horticultural Sciences* **123**: 560–562.

Nerd A, Guttman F, Mizrahi Y. 1999. Ripening and postharvest behaviour of fruits of two *Hylocereus* species (Cactaceae). *Postharvest Biology and Technology* **17**: 39–45.

Nerd A, Tel-Zur N, Mizrahi Y. 2002. Fruit of vine and columnar cacti. In: Nobel PS, ed. *Cacti: biology and uses*. Los Angeles: UCLA Press.

Osman AMA, Reuther W, Erickson LC. 1974. Xenia and metaxenia studies in the date palm *Phoenix dactylifera* L [Pollination experiments]. *Reports of Date Growers Institute* **51**: 6–16.

Purohit AG. 1987. Effect of pollen parent on seed hardness in pomegranate. *Indian Joumal of Agricultural Sciences* **57**: 753–755.

Rahemi M. 1998. Effects of pollen sources on fruit characteristics of 'Shahani' date. *Iran Agricultural Research* **17**: 169–174.

Raveh E, Nerd A, Mizrahi Y. 1998. Responses of two hemiepiphytic fruit-crop cacti to different degrees of shade. *Scientia Horticulturaee* **73**: 151–164.

Sedgley M. and Griffin AR. 1989. *Sexual reproduction of tree crops*. London: Academic Press, 165–167.

Swingle WT. 1928. Metaxenia in the date palm, possibly a hormone action by the embryo or endosperm. *Journal of Heredity* **19**: 257–268.

Tel-Zur N, Abbo S, Bar-Zvi D, Mizrahi Y. 2003. Chromosome doubling in vine cacti hybrids. *Journal of Heredity* **94**: 329–333.

Vashishtha BB. 1986. Abnormal fruiting behaviour of ber (*Ziziphus mauritiana* Lamk.) cultivar Ilayachi. *Annals of Arid Zones* **25**: 165–167.

Wallace HM, Lee LS. 1999. Pollen source, fruit set and xenia in mandarins. *Journal of Horticultural Science and Biotechnology* **74**: 82–86.

Weiss J, Nerd A, Mizrahi Y. 1994. Flowering behaviour and pollination requirements in climbing cacti with fruit crop potential. *HortScience* **29**: 1487–1492.