Comunicata Scientiae
Horticultural Journal

ORIGINAL ARTICLE
published: 29 May 2023
https://doi.org/10.14295/CS.v14.3696

Effects of pollen sources on the quality of nuts borne by pecan cultivars Kiowa and Barton

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Abstract

Selection of pecan cultivars and their pollinizers is essential to reach maximum potential of production. Synchrony of blooming periods of cultivars may not be enough to ensure nut quality because of the xenia effect. This study aimed at evaluating effects of different pollinizers on dimensions and yields of fruit borne by cultivars Kiowa and Barton. Both cultivars were submitted to six treatments. Kiowa: no pollination, free pollination, self-pollination and pollination controlled by pollen from cultivars Cape Fear, Pawnee and Desirable. Barton: no pollination, free pollination, self-pollination and pollination controlled by pollen from cultivars Melhorada, Jackson and Success. Self-pollination decreased dimensions and yields of fruit and kernel. Pollen from different cultivars was found to exhibit positive and negative xenia effect on fruit characteristics. The largest fruit and the highest kernel yield were reached when 'Kiowa' was pollinized with 'Desirable' and 'Barton' was pollinized with 'Success'.

Keywords: *Carya illinoinensis*, cross-pollination, pecan nut tree, self-pollination, xenia

Introduction

Pecan trees are considered allogamous plants that prefer cross-pollination and strongly tend to dichogamy, i. e., to production, viability and dispersion of pollen from staminate flowers, which are non-coincident, either totally (complete dichogamy) or partially (incomplete dichogamy), with receptivity of pistillate flowers (Sparks, 1992; Zhang et al., 2016a).

Even though self-pollination occurs naturally in pecan trees, fruit that originate from cross-pollination are usually larger and have better quality than those that result from self-pollination (Marquard, 1988; Anchondo, 2019). It should also be highlighted that this species exhibits the xenia effect (Romberg & Smith, 1946), which has been defined as the effect of pollen on the development of either endosperm and embryo (seed tissue) or pollen on fruit tissue. It is due to interaction among a nuclear gene of male gamete and two nuclear polar genes of the endosperm or a nuclear gene of the egg, throughout double fertilization in angiosperms (Denny, 1992; Pozzi et al., 2019), and may result in differences in certain characteristics, such as size, shape, color and chemical composition of fruit and seeds (Denny, 1992, Yan et al., 2019).

Some studies have shown the importance of including at least three pollinating pecan cultivars in an orchard to ensure cross-pollination, which leads to high fruit production and quality, since self-pollination results in small and poor-quality nuts (Conner, 2012, Wells 2017). Recommendations are usually based on technical criteria, such as synchronization of pollination with the cultivar of interest, resistance against plagues and diseases and fruit quality. However, information about the effect on the potential to increase fruit yield as the result of pollen sources (xenia) is not given.

Therefore, this study aimed at evaluating the

Received: 21 March 2021
Accepted: 28 March 2022

potential that different pollinizers have to increase quality of fruit borne by cultivars Kiowa and Barton.

Material and Methods

Two experiments were carried out with two cultivars and different pollinizers in two distinct regions to evaluate effects of pollination.

Cross-pollination and self-pollination of the cultivar Kiowa

In this experiment, the cultivar Kiowa was submitted to different pollination treatments. 'Kiowa' plants were implanted in 2010; spacing was 10 x 10 m. They belong to the collection of pecan trees at the experimental station Wilson Ferreira Aldunate, INIA - Las Brujas (Instituto Nacional de Investigación Agropecuaria), in Canelones, Uruguay (34°40'15"S and 56°20'27"W). This cultivar was chosen because of the quality of its fruit and its good adaptation to Uruguayan edaphoclimatic conditions. The experiment was conducted in the 2017/2018 cycle.

Six treatments were used to compose pollination treatments:

1 - No pollination: pistillate inflorescences do not receive any pollen. This treatment was only used to evaluate efficiency of packages in terms of isolation and to monitor fruit drop;

2 - Free pollination: in this treatment, pollination was not manipulated. Inflorescences were only identified to be evaluated.

In controlled pollination, inflorescences were isolated and pollinized with pollen from different cultivars:

3 - Self-pollination: pistillate inflorescences of the cultivar Kiowa were pollinized with their own pollen;

4 - The cultivar Kiowa was pollinized with pollen from 'Cape Fear';

5 - The cultivar Kiowa was pollinized with pollen from 'Pawnee';

6 - The cultivar Kiowa was pollinized with pollen from 'Desirable'.

Cross-pollination and self-pollination of the cultivar Barton

In this experiment, the cultivar Barton, the main one implanted in Brazil, was submitted to different pollination treatments. The study was carried out in the 2018/2019 cycle in a pecan orchard implanted in 2009 in Canguçu, RS, Brazil (31°28'08"S and 52°41'55"W). Spacing was 10 x 10 m.

Six pollination treatments were also used:

1 – No pollination;

2 – Free pollination;

3 - Self-pollination;

4 - The cultivar Barton was pollinized with pollen from 'Melhorada' (Pitol 1);

5 - The cultivar Barton was pollinized with pollen from 'Jackson';

6 - The cultivar Barton was pollinized with pollen from 'Success'.

Treatments were applied to five 'Kiowa' plants and five 'Barton' ones. The pollination treatment was replicated four times in every plant, i. e., four pistillate inflorescences per plant were submitted to the same treatment, totaling 20 replicates.

Pistillate inflorescences (bundles) had to be isolated to prevent pollen blown by the wind from contacting stigmas (except the treatment of free pollination). To carry it out, female inflorescences were bagged in 20 cm long packages, before the stigma became receptive. Collagen casings (7 cm in diameter) were used for the cultivar Kiowa, while cellophane casings (4 cm in diameter) were used for the cultivar Barton. Cotton was wrapped around the base of the branch with the inflorescence and covered with the casing, generating bundles and packages which were then tied with wire. Most little leaves close to the bases of bundles were also bagged and helped to hold the packages.

Pollen of every treatment was collected from staminate inflorescences (aments) at the beginning of the opening of pollen sacs and stored in paper bags under an incandescent lamp so that it could be easily released. It was then collected, sieved and stored in Eppendorf tubes in a freezer at -14 °C up to pollination. In the period of pollen release, it was collected every two days; newly-collected pollen was used to carry out pollination.

Pollination was conducted when stigmas showed bright stigmatic fluid on their surfaces. Pollination processes were carried out with no package removal by a syringe with a hypodermic needle, which was coupled with a rubber bulb by means of a glass tube with a spiral (where the pollen was placed). The needle was inserted through the cotton and directed to the stigmas, where pollen was blown by pressure on the rubber bulb. All inflorescences were pollinized every two days to make sure that all flowers would get pollen when they were receptive. This process was repeated up to the end of the receptivity period of stigmas, when they lost their brightness and got necrotic, condition which was determined by the fact that they got dark.

As soon as stigma surfaces got dark, thus, showing the end of receptivity, packages were removed and the number of nuts per bundle was recorded. Monthly means of nuts per bundle were counted up to harvest.

When capsules (epicarp) that involve nuts opened, thus, showing that they were physiologically mature, harvest was carried out. Afterwards, nuts were dried by a forced air oven at 32 °C (±2 °C) to reach about 4 % moisture, which was determined by the Dickey-John M-3G portable moisture tester.

To determine dimensions and kernel yield in every treatment, 20 fruit samples were randomly separated; every fruit was considered a replicate. Fruit length and width, besides kernel length, width and height and shell thickness were measured by a digital pachymeter. A digital scale, accurate to two decimal places, was used to weigh fruit mass, kernel and shell. In addition, the number of nuts needed to form a kilogram was estimated.

Percentages of increase and decrease as the result of treatments (pollen from different cultivars), by comparison with self-pollination, were also calculated.

In order to meet presuppositions of the analysis of variance, normality and homogeneity of variances were checked by both Shapiro-Wilk and Bartlett tests, respectively. After that, data on evaluations were submitted to the analysis of variance and means were compared by the Tukey's test at 5 % error probability by the SISVAR statistical program (Ferreira, 2011).

Results and Discussion

Inflorescences that did not get pollen (no pollination) exhibited accentuated fruit drop, as expected, reaching 100 % about a month after the blooming period (**Figure 1**). Thus, it may be stated that inflorescences were efficiently isolated, a fact that prevented free pollination from happening.

Nut abscission was found in all treatments during the fruit growth and development phase, but it was more accentuated about 5-6 weeks after pollination (Figure 1). Controlled and free pollination led to means of 4.1 (Kiowa) and 3.5 (Barton) nuts per bundle. They decreased as time went by and totaled 2.6 (Kiowa - Figure 1A) and 1.7 nuts per bundle (Barton - Figure 1B) at the harvest period.

Treatments with pollen from different cultivars did not significantly influence the mean number of nuts per bundle. However, Figure 1B shows that when 'Barton' was pollinized with its own pollen, the percentage of fruit (about 40.0 %) decreased, by comparison with the one of pollination with pollen from other cultivars.

Pollen from different cultivars exhibited significant differences in either nut width or in shell thickness in the case of 'Kiowa'. However, when it was pollinized with 'Desirable' pollen and under free pollination, nut length was significantly higher than the others. Regarding the cultivar Barton, shell thickness was not influenced by different pollen under analysis either. However, when it was pollinized with 'Melhorada', nut width and length were smaller than measures obtained when any other pollen was applied (**Table 1**).

Concerning kernel width, 'Kiowa' had larger dimensions in free pollination conditions which did not differ when the cultivar was pollinized with 'Cape Fear' and 'Desirable'. Kernel length was higher when it was pollinized with 'Desirable', followed by free pollination, with pollen from 'Kiowa' and 'Cape Fear'. Regarding kernel height, there was no significant difference among pollen sources (**Table 2**). It should be highlighted that the area where the experiment of 'Kiowa' pollination was

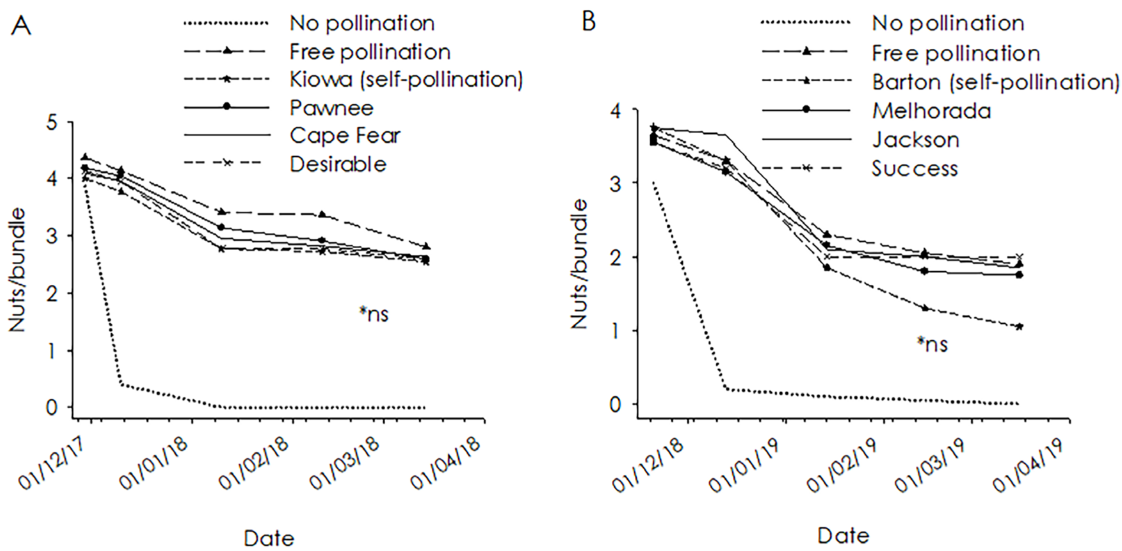

Figure 1. Mean number of nuts per bundle in different pollination treatments (pollen from different cultivars) and evaluation dates of the experiment carried out in Uruguay – A (cultivar Kiowa) and in Brazil – B (cultivar Barton). *ns = not significant at 5 % error probability by the Tukey's test.

Table 1. Width and length of nuts with shell and shell thickness in different pollination treatments (pollen from different cultivars) applied to cultivars Kiowa and Barton

	Nut width		Nut length		Shell thickness	
	mm					
Treatment	Cultivar Kiowa					
Free pollination	20.81	ns	39.94	ab*	0.71	ns
Kiowa (self-pollination)	19.97		39.42	bc	0.73	
Cape Fear	19.73		37.48	c	0.70	
Pawnee	19.58		39.16	bc	0.64	
Desirable	21.38		41.67	a	0.70	
CV (%)	16.81		7.78		14.40	
Treatment	Cultivar Barton					
Free pollination	19.94	ab	38.32	a	0.77	ns
Barton (self-pollination)	19.43	ab	35.18	ab	0.77	
Melhorada	19.28	b	34.37	b	0.70	
Jackson	19.54	ab	36.35	ab	0.72	
Success	20.44	a	37.18	a	0.78	
CV (%)	5.76		6.22		14.40	

*Means followed by the same letter in a column did not differ statistically at 5 % error probability by the Tukey's test; ns not significant; CV = Coefficient of Variation.

Table 2. Kernel width, length and height in different pollination treatments (pollen from different cultivars) applied to cultivars Kiowa and Barton

	Kernel width		Kernel length		Kernel height	
	mm					
Treatment	Cultivar Kiowa					
Free pollination	18.59	a*	31.52	ab	8.05	ns
Kiowa(self pollination)	17.03	b	31.41	ab	7.43	
Cape Fear	17.57	ab	30.77	ab	7.30	
Pawnee	17.25	b	30.29	b	7.61	
Desirable	17.40	ab	33.99	a	7.51	
CV (%)	5.48		8.4		7.44	
Treatment	Cultivar Barton					
Free pollination	15.29	ab	30.14	ns	7.54	ns
Barton(self-pollination)	14.76	ab	28.61		7.64	
Melhorada	14.57	b	28.51		7.58	
Jackson	14.95	ab	30.12		7.47	
Success	15.49	a	30.39		7.53	
CV (%)	5.68		8.55		11.45	

* Means followed by the same letter in a column did not differ statistically at 5 % error probability by the Tukey's test; ns not significant; CV = Coefficient of Variation.

carried out has about 17 cultivars. Several may provide pollen during the receptivity period of stigmas of 'Kiowa', i. e., free pollination was adequate and explained kernel width.

'Barton' pollinized with 'Success' pollen resulted in wider fruit. However, pollination with 'Melhorada' led to the lowest dimension. This cultivar did not exhibit any significant difference in kernel length and height among treatments (Table 2).

Results of free pollination showed that pollen supply for cultivars Kiowa (in Uruguay) and Barton (in Brazil) met the receptivity period of stigmas, since it resulted in mean number of fruit per bundle and dimensions and yield which were similar to the ones of the best cross-pollination treatments (**Table 3**).

Mean fruit and kernel masses of 'Kiowa' were higher in natural pollination conditions, followed by pollination with 'Desirable' and 'Pawnee'. Concerning 'Barton', pollination with 'Success', 'Jackson' and free pollination led to the highest means of fruit and kernel masses (Table 3). It should be highlighted that 'Kiowa' fruit that resulted from pollination with 'Cape Fear' and when self-pollinized were smaller and exhibited lower mean kernel mass, while 'Barton' had lower values when it was self-pollinized and pollinized with 'Melhorada'.

Self-pollination of 'Kiowa' and 'Barton' (even though the latter had no significant difference) decreased percentages of kernel and increased percentages of nut shell (Table 3).

The number of nuts needed to form a kilogram of fruit was significantly influenced by pollination treatments. Regarding 'Kiowa', the lowest number was obtained with free pollination and when it was pollinized with pollen from 'Desirable', while 'Barton' resulted in the lowest number of nuts – to form a kilogram – when pollination was carried out with 'Success' and 'Jackson' and when free pollination was used, since its fruit exhibited the highest mass (Table 3).

Smaller fruit and lower yield were also found when 'Kiowa' was pollinized with 'Cape Fear' and when 'Barton' was pollinized with 'Melhorada'. When pollen from 'Cape Fear' was used to pollinize 'Kiowa', results were lower than the ones found after self-pollination, i. e., there was 2.9 % decrease in fruit mass and 3.5 % more fruit were needed to form a kilogram (Table 3). When 'Kiowa' was pollinized with 'Desirable' and 'Barton' was pollinized with 'Success', there was increase in kernel mass, i. e., 15.5 % and 22.3 %, respectively, by comparison with self-pollination.

According to Díaz (2019), natural abscission of pecan nuts occurs mostly in three periods. The first takes place at the beginning of the development of pistillate flowers since there is no pollination and shortage of plant reserves. The second occurs from five to six weeks after pollination and results from problems caused by pollination, because of inadequate development of the endosperm or no egg fertilization. The third event happens before hardening of fruit shell due to embryo abortion which may be triggered by water or thermal stress or by damage caused by insects. Therefore, the most accentuated abscission found by this study (Figure 1) coincides with the second period, between five and six weeks after pollination. It seems to be the main event of pecan nut abscission, since it not only took

Table 3. Fruit, kernel and shell masses; percentages of Kernel and shell and number of nuts per kilogram in different pollination treatments (pollen from different cultivars) applied to cultivars Kiowa and Barton

	Fruit mass		Kernel mass		Shell mass		Kernel		Shell		Nuts per kg	
	g						%					
Treatment	Cultivar Kiowa											
Free poll.	7.51	a**	4.11	a	3.40	a	54.72	ab	45.28	ab	133	b
Kiowa*	6.68	bc	3.58	b	3.10	ab	53.57	b	46.42	a	150	a
Cape Fear	6.49	c	3.57	b	2.92	b	54.91	ab	45.09	ab	155	a
Pawnee	6.80	abc	3.70	ab	3.10	ab	54.25	ab	45.74	ab	149	a
Desirable	7.42	ab	4.13	a	3.29	a	55.68	a	44.32	b	135	b
CV (%)	6.49		7.25		5.91		1.53		1.84		7.57	
Percentages of increase and decrease with the use of pollination treatments by comparison with self-pollination ('Kiowa')												
Free poll.	12.4		14.8		9.6		2.1		-2.4		-11.3	
Cape Fear	-2.9		-0.4		-5.8		2.5		-2.9		3.5	
Pawnee	1.9		3.4		0.1		1.3		-1.5		-0.9	
Desirable	11.1		15.5		6.1		3.9		-4.5		-10.2	
Treatment	Cultivar Barton											
Free poll.	5.55	ab	2.92	ab	2.63	a	52.37	ns	47.63	ns	183	ab
Barton*	4.99	b	2.63	b	2.37	abc	51.87		48.13		208	a
Melhorada	5.10	b	2.76	b	2.34	c	53.72		46.28		203	a
Jackson	5.29	ab	2.94	ab	2.35	bc	55.55		44.45		191	ab
Success	5.83	a	3.21	a	2.62	ab	54.93		45.07		173	b
CV (%)	13.36		18.03		11.77		8.25		9.57		15.3	
Percentages of increase and decrease with the use of pollination treatments by comparison with self-pollination ('Barton')												
Free poll.	11.1		11.1		11.0		1.0		-1.0		-12.2	
Melhorada	2.1		5.1		-1.2		3.6		-3.8		-2.5	
Jackson	5.9		12.1		-0.9		7.1		-7.6		-7.9	
Success	16.7		22.3		10.5		5.9		-6.4		-16.8	

*Self-pollination; **Means followed by the same letter in a column did not differ statistically at 5 % error probability by the Tukey's test; ns not significant; CV = Coefficient of Variation.

place in pollination treatments, but also occurred in free pollination.

When both cultivars Kiowa and Barton were pollinized with pollen from the cultivars under study, there was no significant effect on the number of nuts per infructescence, a fact that corroborates Ajamgard et al. (2017), who also stated that pollen from different cultivars does not influence the final number of nuts per bundle. Therefore, it may be defined that pecan nut abscission is not only related to pollen from different cultivars but may also be related to certain problems, such as plant nutrition, absence of pollination, inadequate development of endosperm, no egg fertilization, embryo abortion, characteristics of the cultivar itself, temperature, water deficit and damage caused by insects and diseases, according to Sparks (1992), Wood et al. (2010), Wells (2017) and Díaz (2019).

On the same issue, Conner (2007) and Wood (2000) reported that one of the adverse effects of self-pollination is decrease in the number of nuts. However, when cultivars Kiowa and Barton were self-pollinized, they did not exhibit any significant effect on abscission, although the latter showed more accentuated fruit abscission than the former and suggestion of higher fruit abortion when it was self-pollinized (Figure 1).

Besides increase in fruit abscission, self-pollination in pecan trees may also decrease yield in up to 75 % (Conner, 2007; Wood, 2000). Even though results of this study did not reach this level of reduction, it showed that self-pollination of cultivars Kiowa and Barton also had negative effect on fruit size and yield. Marquard (1988) stated that self-pollination decreased fruit mass of the cultivar Western in 20 %, by comparison with 'Western' fruit pollinized with 'Wichita'. It confirms that cross-pollination is one of the factors that enable quality fruit and good yield to be achieved.

It is part of the evolution process of pecan trees, which starts with the mechanism of blooming to avoid self-pollination. According to Conner (2012), in the first step, production of male and female flowers is separated, but it takes place in the same plant (monoecious plant). In the second step, ripening of female and male flowers takes place in different periods in the same plant (dioecious plant). Although Conner (2012) has not described the third step, it seems that it is decrease in fruit quality, so as to diminish self-pollination and, consequently, avoid reproduction of self-pollinized plants. It would happen if both previous steps had "failed", since Conner (2012) reports that self-pollination results in the production of small fruit with low kernel yield. Besides, when progenies are self-pollinized, seedlings tend to be very weak and many die in some years.

Effects of pollen from different cultivars on fruit size has been reported by studies of several fruit trees, such as date palms (Shafique et al., 2011), hazelnut trees (Fattahi et al., 2014), olive trees (Shemer et al., 2014), apple trees (Militaru et al., 2015), grape vines (Sabir, 2015), chestnut trees (Zhang et al. 2016b), camellia (Hu et al., 2020) and pecan trees (Romberg & Smith, 1946, Marquard, 1988; Wang et al., 2010). These studies showed positive and negative effects of pollen on fruit characteristics. It is characterized as the xenia effect, which is quite common in fruit trees.

The xenia effect, which has been described as direct result of pollen on fruit development and characteristics, may influence fruit size, shape and some other characteristics (Denny, 1992; Pozzi et al., 2019), i. e., it is the result of fruit characteristics depending on the cultivar that provides pollen (Denny, 1992; Olfati et al., 2010; Mellizo et al., 2012; Zhang et al., 2016b). This effect may be used to identify the best male progenitor which, once understood, can be used to improve fruit quality.

Results of this study showed that there are positive and negative effects on some characteristics of fruit borne by pecan trees as the result of the cultivar that provides pollen. In other words, different variables of fruit under study were affected at different intensity, depending on the pollen source (Tables 1, 2 and 3). Besides, the xenia effect was observed in both cultivars under study – 'Kiowa' and 'Barton' – produced in Uruguay and Brazil, respectively. Effects of this phenomenon on pecan trees were clear.

Consumers prefer large kernel (Wells, 2017) and this study showed that characteristics may be improved by using cultivars that provide pollen in the receptivity period of stigmas, not only to diminish self-pollination, but also to increase fruit dimensions. It was achieved when 'Kiowa' was pollinized with 'Desirable' and 'Barton' was pollinized with 'Success'. Therefore, results of this study show that attention should be paid not only to the choice of cultivars that can provide pollen in the receptivity period of pistillate inflorescences (pollination synchronization), but also to cultivars which have potential to improve fruit quality. This study shows that the selection of pollinizers has great potential to improve yield and quality of fruit borne by pecan trees.

Conclusion

Self-pollination decreases dimensions and yield of fruit and kernel borne by 'Kiowa' and 'Barton' pecan trees. Pollen from different pecan cultivars exhibited positive and/or negative potential regarding fruit dimensions and yield. 'Desirable' pollinator increases fruit size and kernel yield in the cultivar Kiowa. The same happens when 'Success' pollinizes 'Barton'.

Acknowledgments

To the National Institute of Agricultural Research - Las Brujas to provide the area for study and contribution to carry out the activities. To the National Council of Scientific and Technological Development (CNPq) for the grant of doctoral scholarship of the first author.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.