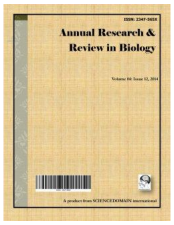

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Supplemental Pollination with Different Sources of Pollen in Olive (*Olea europaea)* 'Manzanilla' under Hot and Arid Environment

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Authors' contributions

This work was carried out in collaboration between all authors. Author GCRL designed the study, wrote the protocol and interpreted the data. Authors GCRL, MDR and RCF anchored the field study, gathered the initial data and performed preliminary data analysis. While authors LCA, MDG and NRF managed the literature searches and produced the initial draft. All authors read and approved the final manuscript.

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ABSTRACT

Aim: Insufficient fruit set is one of the most serious problems that affect the productivity of olive trees in desert area. The objective of this research was to evaluate the supplemental pollination from five different cultivars as sources of pollen for use on 'Manzanilla'.
Materials and Methods: The experiment was carried out in the National Research Institute for Forestry, Agricultural and Livestock, Mexico during 2010. Five sources of pollen ('Barouni', 'Nocellara de Belice', 'Coratina', 'Sevillano' and 'Manzanilla' (self-pollination) were tested. Three

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pollen applications were made (30 g ha^{-1}) for the first application and 60 g ha^{-1} for each of the other two applications.
Results: The germination of pollen grain derivative from the different cultivars was statistically similar, hovewer ranged from 42.1 to 53.5%. There was difference ($P \leq 0.05$) in fruit set, shotberry incidence and olive yield in 'Manzanilla' aftrer supplemental pollination by pollen derivative from diffrent cultivars compared to self-pollinated trees; the yield increased 28%. However, not significant differences were found for fruit set, shotberry or yield among the trees treated with the different sources of pollen, suggesting that 'Manzanilla' is self- incompatible and require cross-pollination to obtain sufficient yield.
Conclusion: Supplemental pollination of 'Manzanilla' olive tree increases fruit yield under hot and arid environment.

Keywords: Olea europaea; cross pollination; shotberry; fruit set; olive yield.

1. INTRODUCTION

Commercial olive production in the world is between 30° and 45° north and south latitude and reaches an annual average of about 12 million tons, of which 90% is to obtain oil and 10% is consumed like table olive. The largest producers of olive are Spain, Italy, Greece and Turkey with the 79% of world production [1] and the global trend of production of olive oil has increased by 97% in the last years and so consumption [2]. Mexico has about 9000 hectares of olive, of which 50% are located in the arid semi-arid regions. Olive production in Mexico is about 30,000 ton per year with a value of about of 21 millions of dollars. By other side, it is estimated that about 60% is dedicated for oil production and 40% for olive table, the main varieties for this purpose are 'Manzanilla' and 'Mission' with 90% and 10% of total acreage, respectively.

The olive tree is a monoecious plant, having both perfect and imperfect flowers [3,4]. Thus, either self-pollination or cross pollination can be found in olive, with wind being the main pollination agent. Traditionally in Spain, olives have been managed as self-compatible and almost no Spanish fields include other cultivars as pollinators [5]. In general olive trees are partly-compatible, but in some climatic conditions various cultivars have problems with pollination and fruit set. 'Manzanilla', one of the most valuable olive cultivars and the one that is most widely cultivated around the world, is self-pollinating, but under hot conditions its pollen tubes grows slowly, resulting in low or no fertilization [3,6].

High temperatures during olive flowering increase pollen incompatibility since pollen tubes frequently become blocked between the stigma and embryo sac when air temperatures increases from 26.7 to 32.2°C. However, pollen tubes from foreign olive cultivars are more suitable, and hence can reach the embryo sacs before they degenerate [7].

Initial olive commercialization effort took place in areas with climates similar to its site of origin, such as in California (USA), Western Australia and Baja California (Mexico). However, in the middle of the twentieth century, commercial plantations began in environments very different from Mediterranean basin, such as the Arid Chaco in Argentina, The Sonoran Desert in the USA and the Altar Desert in Mexico [8]. Early in their life cycle, these olives started to show problems with fruit set and yield, with plant growth and metabolism being profoundly affected by the environmental conditions from these regions [8].

The best Mediterranean olive growing areas are characterized by mild, rainy, winters and long, warm, dry summers [8,9] but extreme temperatures affect the reproductive cycle of the olive [7,8,10]. Cross-pollination is needed when air temperatures during olive bloom exceed 30°C [11] or in years of poor flower quality [5,12,13], to improve fertilization and fruit set. Another problem in warm, dry ecosystems, such as the Arid Chaco of Argentina [8] and Altar Desert in Mexico [14] is very short vernalization period which increases incompatibility of self-pollination for many cultivars including 'Manzanilla', but it has not effect for open-pollination [6].

The objective of this study was to evaluate the supplemental pollination from five different cultivars as sources of pollen for use on 'Manzanilla'.

2. MATERIALS AND METHODS

2.1 Orchard Selection and Management

The experiment was carried out in 2010 year at the National Research Institute for Forestry, Agricultural and Livestock (INIFAP), located in Caborca, Sonora, Mexico (30°42'55''N, 112°21'28''W and 200 m.a.s.l.), in an eleven years old traditional (8x8 m) 'Manzanilla' olive orchard irrigated with 98 cm per year by drip irrigation. The soil was sandy loam with pH 7.96 and electrical conductivity of 1.22 dSm^{-1}. Orchard olive was fertilized with 15-15-15 at rate of 1.5 kg tree (234 kg ha^{-1}) during February and March and with ammonium nitrate (250 kg ha^{-1}) during the postharvest period. The trees were slightly pruned between November 26 and December 21 period. The olive harvest was made manually during first week of August. Other agronomic practices were done in accordance to commercial recommendations [15].

2.2 Pollination Treatments Applied

The trial consisted of supplemental pollination of 'Manzanilla' with four different cultivars of olive as source pollen ('Barouni', 'Nocellara de Belice', 'Coratina', 'Sevillano') and self-pollinated 'Manzanilla'. Three pollen application were made 30 g ha^{-1} for the first application (March 26 when the trees showed 20% of bloom) and 60 g ha^{-1} for each of the two applications (March 29 and April 6, with 60 and 80% of bloom respectively). Pollen application took place between 18:00 and 19:00 hours. Pollen was applied by blower manual along both sides of the olive trees rows. The pollen of four cultivars was manually harvested from trees growing in the Experimental Station. At same time maximum, minimum temperatures, relative humidity and wind velocity data for the applications days were recorded Table 1. Rainfall was not recorded during bloom period.

2.3 Pollen Germination

It was performed as described by [16,17]. Germination medium consisted of 0.8% agar, 10% sucrose, 50 ppm citric acid. The medium was placed in petridishes and sprinkled with olive oil then a small amount of the pollen was spread by a sterilized needle. The petridishes were covered with aluminum foil, and then incubated in chamber room at 27°C for 12 hours. The number of germinated pollen was calculated under light microscope at (10X) magnification power. For each cultivar, three replicates were used. Pollen was considered to be germinated when the length of pollen tube was equal to or exceeded its diameters [18,19].

2.4 Measurement Variables

The parameters evaluated were: 1) fruit set which was obtained by the ratio of fruit number in relation to the number of flowers, 2) the percentage of shotberry that was obtained by the ratio of shotberry fruit to the number of total fruits. Both parameters were made by choosing one floral shoot per each side of the olive trees, 3) yield (kg $tree^{-1}$) and 4) olive fruit quality (diameter, length, weight and pulp to pit ratio) taking a random sample of 100 olive fruits for each tree.

2.5 Statistical Analysis

This study was analyzed using a randomized complete block design, using five treatments with an experimental unit of 15 olive trees. Means were compared by least difference test (LSD) at 5% level of significance. The analysis of variance and means tests were analyzed using the statistical package UANL [20].

Table 1. Pollen germination in four olive cultivars utilized as source of pollen for 'Manzanilla' supplemental pollination

Cultivars	Pollen germination (%)
	Mean ± SD
Barouni	53.5±5.88
Sevillano	51.8±2.59
Nocellara of Belice	51.3±8.15
Coratina	42.1±4.45

3. RESULTS AND DISCUSSION

3.1 Pollen Grains Germination

Pollen germination ranged from 42.1 to 53.5 percent but was similar among cultivars (Table 1). The percentages of pollen germination are similar to those reported by other researchers in 'Sevillano' [21,22], 'Coratina' and 'Frantoio' cultivars [23,24]. According to Griggs et al. [23] these values of pollen germination are adequate for olive. Studies by Mazzeo et al. [25] showed that the germination of the pollen grains of olive was significantly higher in the 'off' year with respect to the 'on' one, which affect the fructification process.

3.2 Fruit Set and Shotberry

Fruit set and shotberry data were taken 80 days after the first pollen application in order to obtain more reliable information, since abscission and other causes affect these traits [4].

There were statistical differences (P≤0.05) between treatments with supplemental pollination versus self-pollination (Table 3). However, the different sources of pollen did not affect fruit set and shotberry. Fruit set of cross pollinated trees varied from 1.9% to 2.4 with 'Barouni' being the highest and 'Nocellara de Belice' and 'Coratina' the lowest. In contrast, the self-pollinated trees had only 0.9% fruit set. The increase in fruit set on 'Manzanilla' with supplemental pollination agree to Grijalva et al. [12,14] and Navarro and Lopez in Mexico [26], and Ayerza and Coates [24] in Argentina and USA. Fruit set in olive is generally from 1 to 3% and this range does not change between 'on' and 'off' years, and is adequate for good production [6,23].

Parthenocarpy produce shotberries but its causes have not been clearly explained. However, it is accepted that profuse numbers of shotberries occur when pollination is poor [3,5]. Shotberry percentage ranged from 5.6 to 9.0 without statistical difference among them, whereas the self-pollination had 15.6 (Table 3). These results are in accordance to those previously reported in 'Manzanilla' [24,25] and 'Kalamata' [27]. Navarro and Lopez, 2013 [26] reported that shotberry under hot and arid environment in Mexico ranged from 5 to 70% on 'Manzanilla', and they concluded that by using 'Sevillano' and 'Barouni' pollen, shotberry was reduced to less than 10%, while self-pollinated trees had 70% shotberry.

Table 2. Maximum and minimum temperature, relative humidity, and wind velocity during pollination of 'Manzanilla' cultivar

Date	Maximum (°C)	Minimum (°C)	Relative humidity (%)	Wind velocity (m/s)
March 26	27.6	9.1	36.3	0.8
March 27	28.3	7.2	32.5	1.1
March 28	31.2	8.2	23.8	0.5
March 29	32.0	6.6	28.9	0.5
March 30	32.3	9.5	23.9	0.6
March 31	28.0	13.3	26.2	2.6
April 1	20.4	8.2	52.3	1.2
April 2	23.9	5.0	52.3	0.4
April 3	28.7	7.7	40.4	0.2
April 4	28.9	10.1	31.3	0.7
April 5	27.1	11.1	37.3	1.1
April6	28.4	7.4	35.2	1.4
April 7	28.4	8.3	24.4	1.4
April 8	29.7	5.5	22.4	0.4

Table 3. Shotberry, percentage of fruit set and yield of olive (*Olea europaea*) 'Manzanilla' after supplemental pollination with pollen derived from different sources

Source of pollen (Cultivars)	Fruit set (%)	Shotberry (%)	Yield (kg tree^{-1})
Barouni	2.4 ^{a}	5.6 ^{a}	60.3 ^{a}
Sevillano	2.1 ^{a}	9.0 ^{a}	67.9 ^{a}
Nocellara de Belice	1.9 ^{a}	6.5 ^{a}	65.4 ^{a}
Coratina	1.9 ^{a}	7.0 ^{a}	63.5 ^{a}
Self-pollination	0.9 ^{a}	15.6 $_{b}$	46.2 ^{b}
Significance	*	*	*

*Means followed by the same letter within columns do not differ significantly (LSD 0.05)*Significant at P≤0.05*

Table 4. Fruit quality of olive (*Olea europaea*) 'Manzanilla' pollinated from different sources of pollen

Source pollen (Cultivars)	Fruit weight (g)	Fruit width (mm)	Fruit length (mm)	Pulp-pit ratio
Barouni	2.93 [a]	1.61 [a]	2.06 [a]	2.69 [a]
Sevillano	3.01 [a]	1.62 [a]	2.05 [a]	2.66 [a]
Nocellara de Belice	3.05 [a]	1.61 [a]	2.04 [a]	2.67 [a]
Coratina	2.93 [a]	1.62 [a]	2.08 [a]	2.70 [a]
Self-pollination	3.07 [a]	1.65 [a]	2.09 [a]	2.68 [a]
Significance	NS	NS	NS	NS

Means followed by the same letter in a column do not differ significantly (LSD 0.05) [NS] Not significant at P≤0.05

3.3 Olive Yield

There were significant differences (P≤0.05) on yield when supplemental pollen was applied in comparison with trees self-pollinated; however, not significant differences in yield among the trees pollinated with different sources. Trees with supplemental pollination had yields from 60.3 to 67.9 kg, respectively (9.41 to 10.60 ton ha^{-1}), while self-pollinated trees had a yield of 46.2 kg (9.24 ton ha^{-1}). The increase in olive production with supplemental pollination was lower compared to other studies conducted in hot and arid environments which report increments from 93 to 98% [12,24]. In this experiment yield was considered excellent as commercial olive orchard since average yield is about 5.0 ton ha^{-1} and varies from 2.0 to 15.0 ton ha^{-1} during 'off' and 'on' years, respectively. High yield obtained in this season could be attributed to optimum temperatures during the time of pollen application (maximum from 27.1 to 32.3°C and minimum from 5 to 11°C), and appropriate relative humidity and wind velocity above (Table 2). For olive trees, optimum flowering occurs when the daily maximum air temperature fluctuates between 16 to 19°C, and the daily minimum is between 2 to 4°C [3]. The need for cross pollination exists when air temperatures during olive bloom exceed 30°C [13]. By the other side, temperatures equal or above 37.8°C during olive bloom reduce fruit set and fruit production [28].

3.4 Olive Quality

Although fruit quality parameters were not statistically affected by treatments, a slight increase on size fruit was found in the self-pollination treatment in comparison to supplemental pollination. Pulp-pit ratio, fruit width and fruit length were not affected by treatment above (Table 4). The differences in fruit quality may be due to the different fruit load among treatments. Griggs et al. [10] indicated that xenia effect has little or no influence on olive fruit development.

4. CONCLUSSION

This study shows that supplemental pollination of 'Manzanilla' olive tree increase fruit yield under a hot and arid environment.

The different source of pollen did not affect the productivity and fruit quality on 'Manzanilla'; however, 'Barouni' it is the most recommended due to the high amount of flowering every year, no alternate bearing and blossoming at the same time that 'Manzanilla'.

Further studies on cross pollination during 'off' year under arid and hot climate are needed.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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