

Determination of Self-Compatibility of the 'Arsel' Olive Cultivar Obtained by Hybridization Breeding

Hükümran GÜL¹  Murat İSFENDİYAROĞLU² 
Nihal ACARSOY BİLGİN²  Rüştü Efe DEĞER³ 

¹Batı Akdeniz Agricultural Research Institute, 07100, Antalya, Türkiye

²Ege University, Faculty of Agriculture, Department of Horticulture, 35100, İzmir, Türkiye

³Departamento de Agronomía, ETSIAM, Universidad de Córdoba, Córdoba, Spain

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Corresponding Author

E-mail: hukumran.gul@tarimorman.gov.tr

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Abstract

This study was aimed to determine the fertilization biology of the 'Arsel' olive cultivar obtained by hybridization breeding. For this reason, treatments of open pollination, cross pollination, and self-pollination were performed for 2 years and self-compatibility and appropriate pollinators of this new cultivar were investigated. Among the varieties included as pollinators ('Memecik', 'Gemlik', 'Uslu', 'Ayvalık' and 'Eğriburun Nizip'), the highest fruit set was achieved with 'Memecik' and 'Gemlik' varieties in both years (2.64%, 1.37% and 1.75%, 1.39%). For this reason, 'Memecik' and 'Gemlik' cultivars were the best pollinator for 'Arsel' olive was thought. On the other hand, considering the statistical analyzes and productivity index values, 'Arsel' variety was found to be self-incompatible. However, the data also indicated that cross pollination was effective in increasing fruit set. Therefore, it is thought that the use of pollinator cultivar in orchard establishment would be beneficial in terms of yield. Pollen viability and germination tests were performed by using 2,3,5 Triphenyl Tetrazolium Chloride (TTC) and agar in petri (15% sucrose + 1% agar + 100 ppm H₃BO₃) methods in the study. The highest pollen viability rate was observed in 'Memecik' cultivar in 2020 and there isn't statistical differences between olive cultivars in 2018. The highest pollen germination rate was observed in 'Arsel' cultivar in 2018. Accordingly, differences were determined between pollen viability and germination rates of examined olive cultivars in terms of years.

1. Introduction

Olive (*Olea europea* L.), which is produced all over the Mediterranean Basin and used as oil and table olives, is one of the most important products of this geography. It is widely cultivated in Türkiye, especially in the Aegean, Marmara and Mediterranean Regions. According to data of the Turkish Statistical Institute (TÜİK), there are almost 192,3 million olive trees with about 1.74 million tons of production in Türkiye. Türkiye ranks fourths in the world in terms of production amount (TÜİK, 2021). It will be possible to have a say in the world market

with the development of new varieties and the spread of production.

Determination of suitable pollinator variety and pollen quality are of great importance in terms of fruit breeding, adaptation and breeding studies. High efficiency ranks first among production targets. For this, first of all, successful pollination and fertilization are required in fruit species. Because low fruit set in olive species is a common problem. Significant production losses are observed due to the absence or insufficient use of the pollinator variety with the main variety. Similarly, self-compatibility of 13 olive species was investigated at

the Olive Land Gene Bank of the 'Olive Research Institute' in Izmir for 2 years. As a result, it was determined that 8 varieties were self-incompatibility, 2 varieties were partially fertile, and 3 varieties were fertile. In addition, it was determined that two hybrid candidates were self-incompatible. As a result, researchers stated that self-incompatibility is common in olives (Gül, 2020). Thus, Mete et al. (2016) reported that although the 'Hayat' olive variety obtained from hybridization studies is self-fertile, it would be appropriate to have a pollinator variety in the orchard in order to increase the fertilization efficiency and fruit set.

There are many studies on the determination of viability and germination of pollen in different olive cultivars. Pollen viability and germination tests are important in fertilization biology studies. It is stated that pollen performances vary according to genotypes and pollen tube development is very slow and does not reach the embryo sac in self-pollination (Porlings and Voyiatsiz, 1976; Palasciano et al., 2008; Selak et al., 2013; Mete et al., 2015).

It is necessary to include 10% pollinator varieties in the orchard, where olive pollen is carried by the wind for very long distances and for an effective pollination. It is reported that this ratio may vary depending on the topography of the region and the wind and air temperature during the flowering period (Lavee and Datt, 1978). In this context, it was determined that the stigma remained receptive for 12 days in the 'Manzanilla' olive variety and the fertilization ratio was 26% in the first 3 days and 55% at the end of the 8th day (Cuevas et al., 2009). In the current study, it was aimed to determine the

appropriate pollinator to be used as a pollinator, self-compatibility and pollen quality of the 'Arسل' olive variety obtained by hybridization breeding by the Olive Research Institute in Izmir.

2. Materials and Methods

The experiment of this study was conducted at the observation plot of registration of Olive Research Institute in Izmir/Kemalpaşa, Türkiye between 2018 and 2020. The study interrupted in 2019, due to strong alternate-bearing tendency of 'Arسل' cultivar. Cultivars like 'Memecik', 'Gemlik', 'Uslu', 'Ayvalık' and 'Eğriburun Nizip' cultivars were used as pollinator for 'Arسل' cultivar. Moreover, open pollination and self pollination applications were conducted. 'Arسل' olive cultivar was developed by hybridization studies and registered on February 2019. This cultivar which showed good performance particularly got with 24.69% oil and 88.24% pulp ratio. Flower clusters were isolated with oily paper bags at the balloon stage of buds to obtain pollens. After the anthesis stage bags were removed and pollens were sieved and protected in the refrigerator in glass bottles. For open-pollination treatments, flowers in clusters, as for the self-pollination, flower buds on clusters take place on one-year-old shoots were counted at balloon stage buds before the flower were counted, isolated and labeled. At the end of the receptivity of pistil, bags were removed and the isolation was completed. The number of flowers in three combinations as open pollination, self pollination and cross pollination was predicted as at least 500 (Figure 1).

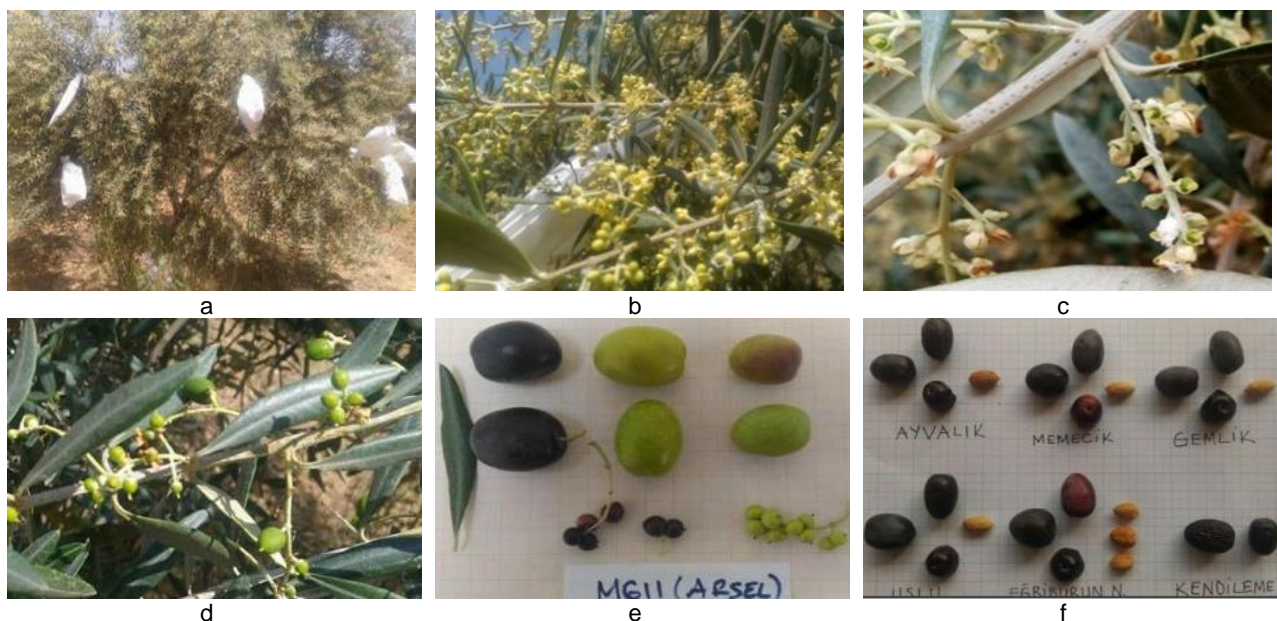


Figure 1. Arsel cultivar (a), beginning of blooming (b), full bloom (c), fruits and unfertilized parthenocarpic fruits 10 days after full bloom (d), fertilized and unfertilized parthenocarpic fruits (e), changes in fruit and seed with the effect of pollinator varieties 3 months after fruit set (f).

Self-fertility of examined cultivars and the degree of fertility of pollinator cultivars were calculated according to productivity index (R) and the obtained figures were evaluated by considering the Table 1 (Moutier, 2002).

$$R = \frac{SP}{OP}$$

where, R is productivity index, SP is percent fruit set obtained from self-pollination or pollinator, and OP is percent fruit set obtained from open-pollination cultivar.

2,3,5 Triphenyl Tetrazolium Chloride (TTC) solution was used to determine the pollen viability rates (Norton, 1966; Heslop-Harrison and Heslop-Harrison, 1970). In this test, which is based on the dyeing of pollen, the red pollen was considered as alive. Unstained pollen was classified as inanimate. In the pollen germination test, in the agar-petri method; 15% sucrose + 1% agar + 100 ppm H₃BO₃ medium was used (Mete, 2009).

The experiment was conducted on a randomized block design with five replications. Variance analysis was applied to the obtained data and Student's T-Test was used to compare the averages.

3. Results and Discussion

The percentages of pollen viability and germination belonging to 'Arsel' olive cultivar, this was used as the main cultivar and the other; pollinator cultivars were given in Table 2.

With the exception of pollen viability rates in 2018, the treatments showed statistically significant differences at ($P < 0,001$) level. The highest rate of pollen viability in 2020 was determined in 'Memecik' (79%) and 'Arsel' (74%) cultivars. These cultivars were followed by 'Uslu' (65%), 'Eğriburun Nizip' (62%), 'Ayvalık' (58%) and 'Gemlik' (49%) cultivars

respectively. In terms of pollen germination rates, the highest figure was determined in 'Arsel' cultivar, while the lowest one was determined in 'Eğriburun Nizip' in the first year of the study. As for the second experiment year, two statistical groups occurred. Accordingly, 'Gemlik', 'Eğriburun Nizip', 'Ayvalık', 'Uslu' and 'Memecik' took place in the first group and 'Arsel' was in the last group.

Pollen viability rates of olive cultivar could be variable, and the possible reasons of this situation might be related to climatic factors, cultural practices, tendency to alternate-bearing together with genetic factors as formerly exposed in different studies (Ferri et al., 2008; Palasciano et al., 2008; Mete., 2009; Gierdani et al., 2012; Mete et al., 2012; Manzzeo et al., 2014; Abacı and Asma, 2015; Karabıyık and Eti, 2015).

Pollen viability values were higher than germination values in this study which were with the former results of Rovira and Tous (2002). One of the possible reasons of this situations, despite the acceptance of the viable pollens have a good germination capacity, as a result of the insufficient in-vitro conditions for pollen germination, lower germination rates could be generally be obtained as formerly suggested by Yıldız and Kaplankıran (2014). Similar result was emphasized in various stone fruit species (Eroğlu and Mısırlı, 2016). Moreover, in 'Arsel' cultivar which was determined in self-incompatible category in accordance with pollination biology in this study, obtained pollen germination rate as 19% in 2020 might be one of the possible causes of self-incompatibility was thought. Accordingly, the low pollen germination rate can be resulted with the decreases in germination and fruit set consecutively.

The fruit set and productivity index (R) values related with the combinations of controlled pollinations that conducted in 'Arsel' cultivar, which was used as the main cultivar have seen in Table 3. Statistically significant ($P < 0,001$) differences were predicted between treatments in both years (2018-

Table 1. Classification of self-fertility and activity level of pollinators.

(R)	0.00	0.15	0.15	0.30	0.30	1.00
	Self-incompatibility		Partially self-compatibility		Self-compatibility	
(R)	0.00	0.33	0.33	0.66	0.66	1.00
	Bad pollinator		Passable pollinator		Good pollinator	

Table 2. Pollen viability and germination ratio (%).

Cultivars	Pollen viability ratio		Pollen germination ratio	
	2018	2020	2018	2020
Arsel	89.82	74.00 ab	64.28 a	19.00 b
Memecik	93.05	79.00 a	36.47 c	26.00 a
Uslu	78.37	65.00 bc	35.44 c	29.00 a
Eğriburun Nizip	87.60	62.00 c	24.12 d	37.00 a
Ayvalık	84.01	58.00 c	34.89 c	35.00 a
Gemlik	81.93	49.00 d	45.69 b	37.00 a
CV%	1	9	2	6

Means were grouped according to Student's t test ($P < 0,001$).

2020) and data were in parallel with normal separation. Fruit set rate of 1-2% in olive was accepted as sufficient by different researchers. The combination in which the 'Memecik' cultivar was used as pollinator for 'Arsel' cultivar, fruit set rates were 2.64% and 1.37% respectively. In terms of Number of Fruits Per Inflorescence (NFPI) in 2018, 'Memecik' cultivar was ranking first and it was followed by 'Gemlik', open pollination, 'Ayvalık', 'Uslu', 'Eğriburun Nizip' and self-pollination respectively. According to Productivity Index (R) values of 2018, while 'Memecik', 'Gemlik', 'Ayvalık' and 'Uslu' cultivars were in good pollinator class, 'Eğriburun Nizip' took place in acceptable pollinator class. Moreover, 'Arsel' cultivar was classified as self-incompatible. In terms of NFPI in 2020, 'Memecik', 'Gemlik' and open pollination took place in the same statistical group in both years and gave the highest fruit set values as well ($P < 0,001$). According to Productivity Index (R) values, while 'Memecik' and 'Gemlik' cultivars in good pollinator class, 'Ayvalık' took place in acceptable pollinator class. On the contrary, 'Uslu' cultivar was in bad pollinator class as it was calculated. 'Arsel' cultivar was also self-incompatible like the first year.

In Kemalpaşa location, blooming period was observed between April 27 and May 15 in 2018. As the start of blooming in April 27 and May 3-4 when the full blooming was observed in 'Arsel' cultivar, daily maximum temperatures ranged between 28-33°C and these values did not cause any problem related with pollination biology was determined (Figure 2).

Moreover, after quite low precipitations measured as 16 and 5 mm, on May 6 and 11, resulted with proper conditions for pollination biology was thought. In fact, results were quite satisfactory clue to fruit setting values and 2,64% fruit setting was determined in combination, in which 'Memecik' cultivar was used as pollinator and more than 1% fruit settings were also obtained with the other combinations (Table 3).

In 2020, daily maximum temperatures were higher than 35°C at the start of blooming (May 14) and during full blooming (May 17-18) have seen in Figure 3.

However, these high temperatures, which may adversely affect the fruit set, dropped after the precipitation occurred on May 21. After that, lower daily maximum temperatures were measured to the

Table 3. Fruit set rates depending on pollination combinations in 'Arsel' female parent.

Application	Fruit set ratio (%)		NFPI		Productivity index (R)	
	2018	2020	2018	2020	2018	2020
Memecik	2.64 a	1.37 a	0.43 a	0.17 a	1.00	1.00
Gemlik	1.75 ab	1.39 a	0.28 b	0.17 a	1.00	1.00
Uslu	1.17 bc	0.42 b	0.17 bc	0.04 b	0.73	0.28
Ayvalık	1.11 bc	0.46 b	0.19 bc	0.05 b	0.82	0.35
Eğriburun Nizip	0.83 c	-	0.10 cd	-	0.43	-
Open pollination	1.47 bc	1.08 a	0.23 bc	0.14 a	1.00	1.00
Self-pollination	0.15 d	0.13 b	0.02 d	0.01 b	0.08	0.07
(CV, %)	19	21	8	5		

NFPI: Number of fruits per inflorescence. The means were classified according to the original values by Student's t test, and the groups were classified according to the transformed values ($P < 0,001$). The data fit the normal distribution.

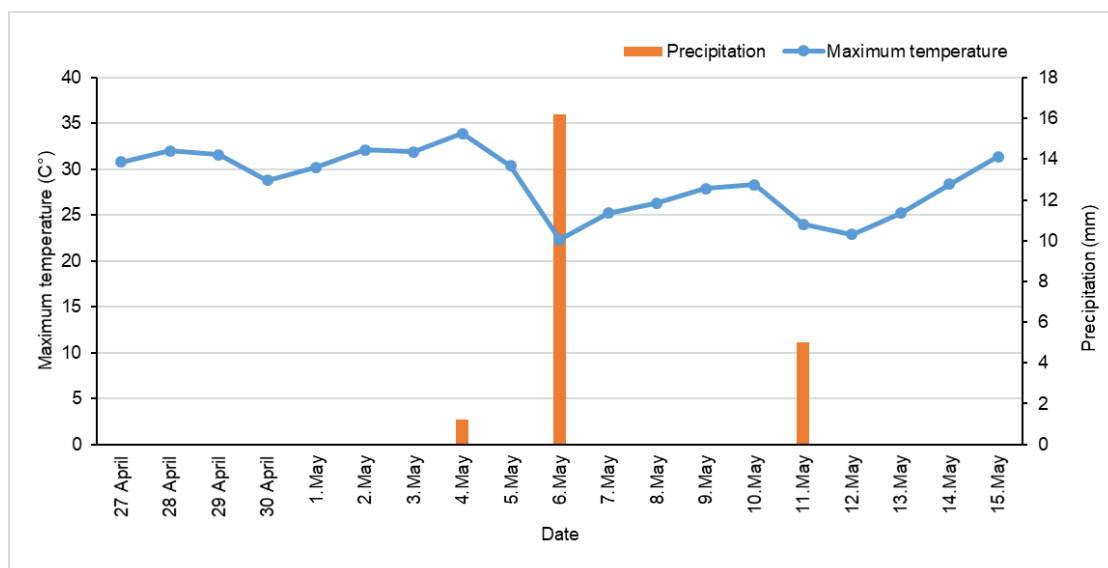


Figure 2. The daily maximum temperature and precipitation in the blooming period of 2018.

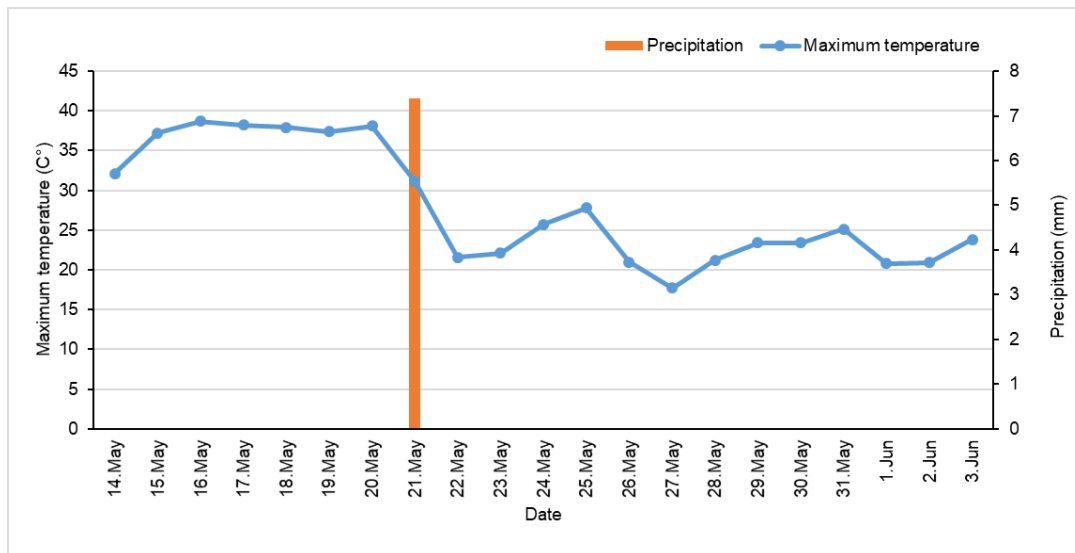


Figure 3. The daily maximum temperature and precipitation in the blooming period of 2020.

end of blooming (June 3), which gave rise to quite proper conditions for pollination biology was observed. Considering the all combinations of the second pollination were done after the precipitation on May 21, in terms of fruit set rates obtained as 1,37% from 'Memecik' and 1,39% from 'Gemlik' combinations were quite good figures was thought. Fruit set at 1-2% have been accepted as sufficient for olive, in general. In both years of the study, in accordance with the pollinator cultivars, quite satisfactory fruit set rates occurred. However, when the meteorological data were considered, fruit set rates obtained in 2018 were higher compared with 2020, due to the more proper daily maximum temperature values. Moreover, a difference of 10-17 days between inflorescence initiation and blooming was determined in relation with the warmer weather conditions during blooming period in 2018.

4. Conclusion

For 'Arsel' olive cultivar, 'Memecik' and 'Gemlik' cultivars were the combinations that provided the highest level of fruit sets related either with fruit number per inflorescence, which is basic for evolution, or values of productivity index (R). For this reason, mentioned cultivars were the proper pollinator for 'Arsel' cultivar olive. Moreover, higher fruit set was obtained compared with open pollination treatment. Plantations of 'Arsel' should include a minimum of 10% of trees of 'Memecik' or 'Gemlik' in order to ensure a good compatible pollination. According to this study, for 'Arsel' cultivar which was obtained with the hybridization of 'Gemlik' and 'Memecik' cultivars, determination of parent cultivars as the most proper pollinator should be evaluated as a quite interesting result for olive breeding programs.

References

- Abacı, T.Z., & Asma, M.B. (2015). Pollen vitality, germination conditions and pollen tube length investigation of hybrid apricot genotypes. *Anadolu Journal of Agricultural Science*, 29(1):12-19.
- Cuevas, J., Pinillos, V., & Polito, S. (2009). Effective pollination period for 'Manzanillo' and 'Picual' olive trees. *Journal of Horticultural Science & Biotechnology*, 84(3):370-374.
- Eroğlu, Z., & Mısırlı, A. (2016). The determination of pollen quality in some peach cultivars and types. *Ege University Agriculture Faculty Journal*, 53(1): 83-88.
- Ferri, A., Giordani, E., Padula, G., & Bellini, E. (2008). Viability and in vitro germinability of pollen grains of olive cultivars and advanced selections obtained in Italy. *Advances in Horticultural Science*, 22(2):116-122.
- Gierdani, E., Ferri, A., Trentacoste, E., & Radice, S. (2012). Viability and in vitro germinability of pollen grains of olive cultivars grown in different environments. *VII International Symposium on Olive Growing Acta Horticulture*.1057.5.
- Gül, H. (2020). Bazı zeytin çeşitlerinin kendine verimlilik durumlarının saptanması. MSc Thesis, Ege University, 48 p (in Turkish).
- Heslop-Harrison, J., & Heslop-Harrison, Y. (1970). Evaluation of pollen viability by enzymatically induced fluorescence. Intracellular hydrolysis of fluorescein diacetate. *Stain Technology*, 45(3):115-120.
- Karabıyık, Ş., & Eti, S. (2015). Determination of pollen viability, germination levels and amount of pollen production of some loquat cultivars at different flowering periods. *Fruit Science Journal*, 2(1):42-48.
- Lavee, S., & Datt, Z. (1978). The necessity of cross pollination for fruit set of Manzanillo olives. *Journal of Horticultural Science*, 53: 61-266.
- Manzreo, A., Palasciano, M., Gallotta, A., Camposeo, S., Pacifico, A., & Ferrara, G. (2014). Amount and quality of pollen grains in four olive (*Olea europaea* L.) cultivars as affected by 'on' and 'off' years. *Scientia Horticulturae*, 170(7):89-93.
- Mete, N. (2009). Bazı Zeytin çeşitlerinin Döllenme Biyolojisi Üzerinde Araştırmalar. MSc Thesis, Ege University, 21 p. (in Turkish).

- Mete, N., Mısırlı, A., & Çetin, Ö. (2012). Determining the biology of fertilization and pollinators in some olive cultivars. *Proceedings of the 4th International Conference on "Olive Culture and Biotechnology of Olive Tree Products"*. 69-74 pp.
- Mete, N., Şahin, M., & Çetin, Ö. (2015). Determining of pollen viability and germination in some olive cultivars. *Olive Science*, 5(1):9-12.
- Mete, N., Şahin, M., & Çetin, Ö. (2016). Determination of self-fertility of the 'Hayat' olive cultivar obtained by hybridization breeding. *Journal of Tekirdag Agricultural Faculty*, 2016:13.
- Moutier, N. (2002). Self-fertility and inter-compatibilities of sixteen olive varieties. *Acta Horticulturea*, 586.
- Norton, J.D. (1966). Testing of plum pollen viability with tetrazolium salts. *Proceedings of the American Society for Horticultural Science*, 89:132-4.
- Palasciano, M., Camposeo, S., Ferara, G., & Godini, A. (2008). Pollen production by popular olive cultivar. *Acta Horticulturea*, 791:489-492.
- Porlingis, I.C., & Voyiatzis, D., (1976). Effect of growth substances on fruit-set in a partly self-incompatible olive cultivar. *Journal of the American Society for Horticultural Science*, 101:432-434.
- Rovira, M., & Tous, J. (2002). Pollen viability in several 'Arbequina' olive oil clones. *Acta Horticulturea*, 586:197-200.
- Selak, G.V., Perica, S., & Poljak, M., (2013). The effect of temperature and genotype on pollen performance in olive (*Olea europaea* L.) *Scientia Horticulturae*, 156(7):38-46.
- TÜİK, 2021. Türkstat, Crop Production Statistics. <http://tuikapp.tuik.gov.tr/bitkiselapp/bitkisel.zul>, Access Date: 08.02.2021.
- Yıldız, E., & Kaplankıran, M. (2014). The pollen viability and germination ratio of different persimmon genotypes. *Ege University Agriculture Faculty Journal*, 51(2):117-123.