

Comparative study on *in vitro* micropropagation response of seven globe artichoke [*Cynara cardunculus* var. *scolymus* (L.) Fiori] cultivars: open-pollinated cultivars vs F₁ hybrids

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Abstract

Globe artichoke [*Cynara cardunculus* var. *scolymus* (L.) Fiori] growing has gained commercial importance in recent years due to its consumption as food. It has also started to attract attention in pharmaceuticals. Due to globe artichoke's stated importance, growers need large amount of pathogen-free, healthy starting materials for production. Stated material will maximize the yield while minimizing the costs. Hybrid cultivars have uniform in height and maturity and could be harvested concurrently; on the other hand, an open-pollinated cultivar would have useful potential that could be smoothly produced locally at a lower cost. *In vitro* micropropagation enabling these goals as it serves large scale, fast, reliable and realistic alternative method to classic propagation via offshoots. The aim of the present study was to comparatively evaluate the micropropagation efficiency of two important local open-pollinated (OP) cultivars ('Bayrampaşa', 'Sakız') and five F₁ hybrid cultivars ('Olympus', 'Madrigal', 'Sambo', 'Green Globe', 'Imparator'), on the basis of total subcultures they were subjected to. Various plant growth regulators at various combinations were assessed for *in vitro* micropropagation and subsequent *in vitro* rooting. 3/4 basic MS medium supplemented with 0.05 mg L⁻¹ BA + 0.005 mg L⁻¹ IBA was determined as the best media combination for *in vitro* micropropagation, while 10.0 mg L⁻¹ IAA + 1.0 g L⁻¹ activated charcoal adding to 1/2 basic MS medium had positive effects on *in vitro* rooting. According to results, the micropropagation efficiency varied based on cultivar differences and number of subcultures regardless of being OP or F₁ hybrid. The present study demonstrated that *in vitro* propagation of globe artichoke could be a valuable process for assessing mass propagation regardless of using F₁ or OP cultivars. Considering the OP cultivars are cheap in terms of price in a comparison to F₁ hybrid cultivars, OP cultivars could be also recommended to be used for *in vitro* mass propagation.

Keywords: globe artichoke; *in vitro* response; *in vitro* mass propagation; plant growth regulators; plant tissue culture

Introduction

Globe artichoke [*Cynara cardunculus* var. *scolymus* (L.) Fiori], which is an important crop and alternative medicine, has been grown intensely, especially in the Mediterranean basin countries. It has grown

for its immature flowers (heads) has recently begun to be used in many fields, especially in pharmaceuticals, due to the high bioactive components of its body and leaves which are still being considered as waste material.

It can be produced both sexual and asexual. Sexual production is provided by seeds, asexual production refers to vegetative production. It is a cross-pollinated plant species and when propagated by seed, plants display high heterogeneity. Due to that it is generally not preferred for commercial production. Traditionally, vegetative propagation has become prominent (Bekheet *et al.*, 2014). Offshoot or ovoli is used for vegetative propagation. However, vegetative propagation has some difficulties such as transferring some diseases and pests through the main plants used in the reproduction, the low reproduction rate during the growing period hinders genetic and agronomic development, physiological differences between shoots taken from the same parent plant and being less suitable for the mechanization (Ancora, 1986; Ordás *et al.*, 1990; Morzadec and Hourmant, 1997; Rey *et al.*, 2013; Saccardo *et al.*, 2013; Bekheet *et al.*, 2014; Campanelli *et al.*, 2014; Dawa *et al.*, 2018; Mazzeo *et al.*, 2020).

In vitro methods help to overcome such problems and are seen as fast, reliable and realistic alternative methods that can be used for large scale production (Rey *et al.*, 2013; Campanelli *et al.*, 2014). Among these *in vitro* methods, the micropropagation technique was first developed in globe artichokes by De Leo and Greco (1973), and is a powerful alternative to traditional propagation. Homogenous, free from diseases and pests, healthy plants can be obtained by *in vitro* micropropagation method, while this method also provides a high reproduction rate. In previous micropropagation studies shoot tip, meristem and seeds were used as explants (Ancora *et al.*, 1981; Pécaut *et al.*, 1983; Rossi and De Paoli, 1992; El Boullani *et al.*, 2012; Bekheet *et al.*, 2014; Dawa *et al.*, 2018). Due to the infection problem encountered in shoot tip culture, researchers decided to use meristem culture in order to obtain healthy, virus and other pathogens free plantlets. Different cultivars need different kind of incubation practices as well as different media combination and plant growth regulators. If all stated parameters are optimized, healthy plants can be obtained and the micropropagation rate can be increased. Plants obtained by *in vitro* micropropagation method perform well in terms of both qualitative and quantitative properties in cultivation and therefore compensate high *in vitro* production costs (Saccardo *et al.*, 2007). In addition to all of these positive properties, it is important to provide an opportunity for the selection of plants with the high medical value among plants that are rapidly propagated *in vitro*.

Growers believe that the key to success requires combination of various approaches. Due to F₁ hybrids supplying a high degree of uniformity, disease resistance, and increased yield and "hybrid vigor", F₁ hybrids are preferred by the majority of growers (Bonasia *et al.*, 2010; Saccardo *et al.*, 2013). On the other hand, it is also known that open-pollinated (OP) globe artichoke cultivars are prominent in some countries in the world as they are ideally adapted to certain environmental conditions. Due to the fact that OP's higher degree of genetic heterogeneity, they have high genetic diversity (Noorani *et al.*, 2013; Mauromicale *et al.*, 2018). Diminishing the possibility of disease or other genetic problems may help to protect from the environmental stresses. Additionally, it can be identified as a new trend that promoting some OP cultivars use among growers is encouraged by several governments in developing countries.

As stated above several *in vitro* studies were conducted on *in vitro* micropropagation of globe artichoke. To our knowledge, there is no study comparing the *in vitro* micropropagation responses of OP cultivars and F₁ hybrids in globe artichoke. The present study was therefore conducted to reveal the *in vitro* micropropagation responses of 2 important local OP globe artichoke cultivars and 5 commercial F₁ hybrid globe artichoke cultivars.

Materials and Methods

Plant material

In the present study, two important local OP globe artichoke cultivars, namely 'Bayrampaşa' and 'Sakız' and five F₁ hybrid globe artichoke cultivars namely 'Olympus', 'Madrigal', 'Sambo', 'Green Globe' and 'Imparator' were used as plant materials.

Culture preparation stage

For the initiation of micropropagation, young shoots, about 10-15 cm size, were used as explant. The shoots collected from the experimental field were first washed under running tap water and soak in antibacterial soap for 10 minutes (900 mL of purified water + 100 mL of antibacterial soap). Subsequently, these plant materials were subjected to 2-stage surface sterilization in a laminar air flow workbench. In the initial surface sterilization process, the plant materials were treated with 40% (4.5% sodium hypochlorite, v/v) commercial bleach solution for 15 minutes and then rinsed 3 times with sterile distilled water. After the first stage of surface sterilization, the plants' surface areas were reduced by cutting. Second surface sterilization stage was applied to these plant materials with 20% of commercial bleach solution (4.5% sodium hypochlorite, v/v) for 5 min and rinsed 3 times with sterile pure water. After successful 2-stage sterilization procedures, the meristems were separated from the main plant material by sterile forceps and scalpel and cultured in the medium prepared for micropropagation (Figure 1).

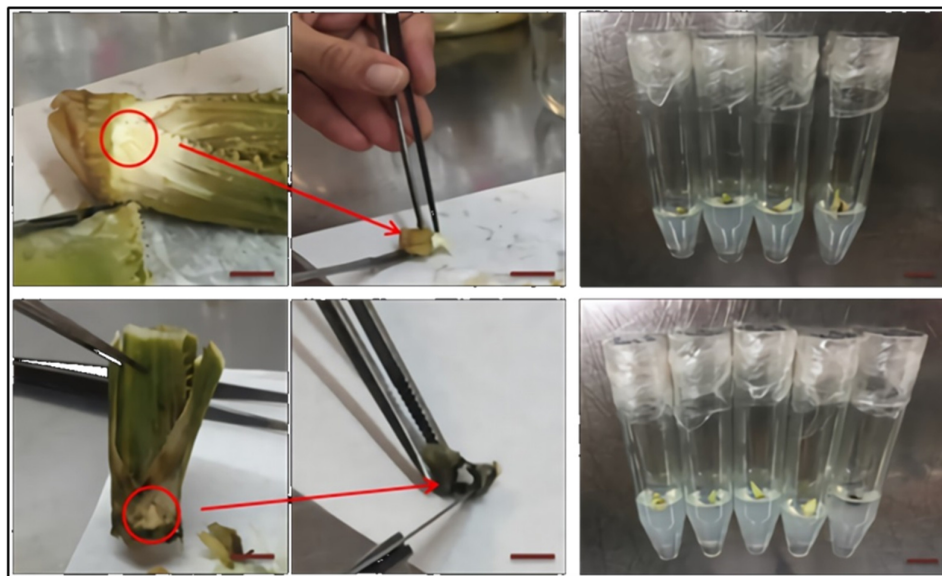


Figure 1. Separation of meristems from mother plant explants and culturing in micropropagation medium (bar = 1.0 cm)

Murashige and Skoog (MS) (1962) was used as the basic medium for micropropagation. Two different media combinations were used in the study: $\frac{3}{4}$ MS medium was supplemented with 0.05 mg L⁻¹ BA + 0.005 mg L⁻¹ IBA (No.1) and 0.25 mg L⁻¹ BA + 0.025 mg L⁻¹ IBA (No.2). These two media combinations were combined with sucrose (3.0%) and agar (6.0 g L⁻¹) and pH was adjusted to 5.8 before autoclaving.

Meristem culture induction and axillary shoot proliferation

The meristems were initially cultured in test tubes containing 10 mL of medium with one explant per tube (Figure 2). After 4 weeks, the shoots of the meristems were cut off to 1-3 cm length and transferred to 500 mL glass jars containing 100 mL medium. During subcultures, 5 explants were placed in each jar. Sub-culturing

was repeated with 4 weeks intervals and continued until the 10th subculture. Cultured plants were kept under 25±1 °C and 16/8 photoperiod conditions.

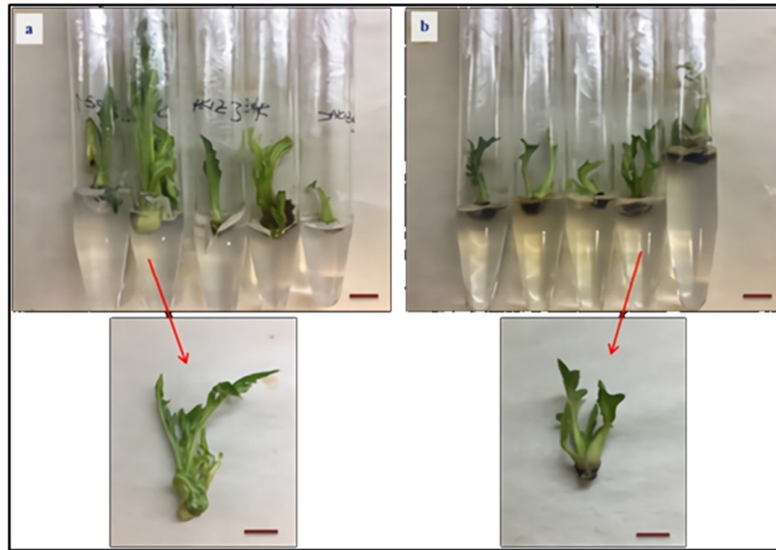


Figure 2. Observation of 1 month after the meristems was cultured; a. Sakız OP, b. Olympus F₁ (bar = 1.0 cm)

In vitro root induction and acclimatization

For root induction, various media combinations were assessed. To serve the purpose, indole-3-acetic acid (IAA, 10.0 mg L⁻¹), indole butyric acid (IBA, 6.0 mg L⁻¹), gibberellic acid (GA₃, 5.0 mg L⁻¹), and naphthalene acetic acid (NAA, 0.5 mg L⁻¹) were used at different concentrations and combinations with ½ basic MS medium (Ozsan and Onus, 2019). The concentrations of plant growth regulators and activated charcoal (AC) doses (1.0 g L⁻¹ and 2.0 g L⁻¹) used in present study were based on previous studies (Ancora *et al.*, 1981; Morzadec and Hourmant, 1997; Cavallaro *et al.*, 2004; Tavazza *et al.*, 2004; López-Pérez and Martínez, 2015; Ercan, 2016).

While the micropropagation was in progress, the acclimatization of *in vitro* rooted plantlets to the *in vivo* conditions was also carried out. For this purpose, *in vitro* rooted plantlets (Figure 3) were transplanted into plastic trays containing peat: perlite (1:1) mixture. Plastic trays were covered with polyethylene cover during 10 days. The plantlets were watered twice a day. During 30-40 days of acclimatization process observations were made on plantlets and at the end acclimatization period surviving plants were transferred to the field.



Figure 3. Rooted plantlets belonging to Sakız OP cultivar (bar = 1.0 cm)

Evaluated parameters

In vitro micropropagation rates, the average number of axillary shoots per explant, the average number of leaves per axillary shoot, *in vitro* root formation and respond to acclimatization were compared on the basis of being OP and F₁ hybrid globe artichoke cultivars. Since each subculture was carried out with 4 weeks intervals, the number of new shoots was counted to calculate multiplication rate (MR) at the end of 4 weeks.

Statistical analysis

The experiment was performed in three replications for comparing 2 OP globe artichoke cultivars and 5 F₁ hybrid globe artichoke cultivars to analyze their *in vitro* micropropagation response. Three jars were used in each replicate and 5 explants were used in each jar. The data obtained were subjected to variance analysis in the JMP package program and the differences between the averages were determined by LSD test.

Results and Discussion

Disinfection protocol

Globe artichoke can be propagated via *in vitro* methods or traditional methods by using offshoots. In a comparison between *in vitro* propagation and traditional propagation methods, *in vitro* propagation has several advantages such as high multiplication rate and obtaining disease free starting material. But *in vitro* multiplication methods have its own disadvantages, too i.e. infection; shoot hyperhydricity and browning (Alp *et al.*, 2010).

This is why the plant materials were subjected to 2-stage surface sterilization. The initial disinfection stage was carried out with high concentration commercial bleach to remove soil-based infection/contamination sources. After the 1st stage, the plant materials were trimmed down to make them ready for 2nd stage disinfection process in order to prevent further infection/contamination. Since the results obtained from present study are highly positive regarding disinfection, 2-stage surface sterilization can be recommended for future studies.

Evaluations of media combinations

The micropropagation was conducted with two media combinations until 3rd subculture. In previous studies, it was stated that MS basic medium supports *in vitro* shoot formation in globe artichoke (De Leo and Greco, 1973; Ancora *et al.*, 1981; Pécaut *et al.*, 1983; Iapichino, 1996; Alp *et al.*, 2010; El-Zeiny *et al.*, 2013; López-Pérez and Martínez, 2015; Dawa *et al.*, 2018). Throughout the experiments, media combinations were carried out with MS basic medium and it was found that MS basic media supplemented with 0.05 mg L⁻¹ BA + 0.005 mg L⁻¹ IBA combination (No.1) was more successful than media combination of 0.25 mg L⁻¹ BA + 0.025 mg L⁻¹ IBA (No.2) in terms of general appearance, growth and development of plantlets. It is why all the plantlets originating from media combination No.2 were discarded and plantlets coming from media combination No.1 were used for the rest of the experiment period. Results on media evaluation clearly showed that medium No.1 was superior over medium No.2. In other words, lower concentrations of BA and IBA resulted with good growth and development of *in vitro* plantlets of the all globe artichoke cultivars used. Similar findings were also reported in previous studies. In a study conducted by Bedini *et al.* (2012), they reported that combination of BA and IBA at reduced concentrations was very successful for *in vitro* propagation of globe artichoke. Several studies also reported that the use of low plant growth regulators concentrations promoted and enhanced the plantlet quality (Ancora *et al.*, 1981; Pécaut *et al.*, 1983; Rossi and De Paoli, 1992; Morone Fortunato and Ruta, 2003; Castiglione *et al.*, 2007; Bedini *et al.*, 2012). It is assumed that increasing plant growth regulator concentration stimulate negative symptoms such as browning and shoot hyperhydricity (Rossi and De Paoli, 1992; Brutti *et al.*, 2000; Pacifici *et al.*, 2007; Bedini *et al.*, 2012). So, findings of the present study are in accordance with previous studies.

Multiplication rates (MR)

There was no rapid increase in MR at the beginning of culture for OP and F₁ hybrid globe artichoke cultivars. As the number of subcultures increased, the shoots became well adapted to *in vitro* conditions and MR, therefore, increased (Table 1). Many researchers think that low MR at the beginning of culture is associated with the initial stress of shoots and as a result of getting adapted to *in vitro* conditions in due time either MR increases or inter subcultural fluctuations are minimized (Phillips *et al.*, 1994; Zucchi *et al.*, 2002; Rey *et al.*, 2013). Findings of present study about MR are, thus, in agreement with previous studies. MR showed variation based on cultivar being OP or F₁ hybrid and as well as number of subcultures. ‘Bayrampaşa’ OP globe artichoke cultivar and ‘Madrigal’ and ‘Imparator’ F₁ hybrid globe artichoke cultivars reached the maximum MR at the 10th subculture. On the other hand, ‘Green Globe’ and ‘Sambo’ F₁ hybrid globe artichoke cultivars were reached to maximum MR at the 8th subculture, while ‘Sakız’ OP globe artichoke cultivar and ‘Olympus’ F₁ hybrid globe artichoke cultivar reached to maximum MR at the 6th subculture. Although the ‘Bayrampaşa’ OP and ‘Imparator’ F₁ hybrid globe artichoke cultivars reached the maximum MR in the 10th subculture, these values were found to be the highest MR values when the overall study was evaluated. Although, Madrigal F₁ hybrid cultivar reached the maximum MR in the 10th subculture like these cultivars, the MR of ‘Bayrampaşa’ OP and ‘Imparator’ F₁ hybrid globe artichoke cultivars were about 5 times higher than ‘Madrigal’ F₁ hybrid cultivar. Although, ‘Sakız’ OP cultivar reached the maximum MR in the 6th subculture there were fluctuations between the 7th and 10th subcultures. The most fluctuations on MR values were recorded in F₁ hybrid cultivars, ‘Madrigal’, ‘Sambo’ and ‘Green Globe’. In previous studies, it was revealed that there was a relationship between the number of subcultures and *in vitro* proliferation. El-Zeiny *et al.* (2013) reported that they recorded an increase in *in vitro* production until the 5th subculture in a total of 7 subcultures conducted in their study. Similarly, Dawa *et al.* (2018) stated that when the number of subcultures was increased *in vitro* multiplication rate also increased until a certain number of subcultures. It is thought that decision of how many subcultures needed to reach to certain number of plants should be done heavily depends on the cultivars used for *in vitro* micropropagation.

Table 1. Multiplication rates/subculture based on cultivars (%)

No of sub-culture	Cultivars						
	‘Bayrampaşa’ OP	‘Sakız’ OP	‘Olympus’ F ₁	‘Madrigal’ F ₁	‘Sambo’ F ₁	‘Green Globe’ F ₁	‘Imparator’ F ₁
0	3.00 p-u	1.33 uv	1.66 tuv	4.33 n-t	4.66 m-s	5.33 l-p	6.66 j-n
1	4.33 n-t	3.33 o-u	1.66 tuv	3.00 p-u	3.00 p-u	2.33 r-v	5.00 l-r
2	7.66 jkl	6.00 k-o	2.00 s-v	2.33 r-v	1.33 uv	3.66 o-u	9.00 hij
3	28.00 e	25.33 e	7.33 j-m	3.33 o-u	1.66 tuv	8.33 ijk	33.33 d
4	76.00 w	86.00 v	26.33 e	6.66 j-n	1.00 uv	33.66 d	76.00 w
5	199.66 o	256.00 l	75.00 w	10.66 ghi	11.33 gh	61.00 z	94.66 u
6	457.00 f	512.66 e	226.66 n	21.00 f	12.00 g	38.00 c	154.66 p
7	652.66 d	312.00 j	144.00 q	44.00 b	85.00 v	35.66 cd	230.33 m
8	697.66 c	300.00 k	111.66 st	109.33 t	113.33 s	62.00 yz	421.33 h
9	700.00 bc	256.00 l	146.00 q	65.00 x	64.00 xy	22.00 f	442.66 g
10	700.66 b	345.00 l	84.33 v	134.00 r	56.33 a	0 v	709.66 a
Averages of cultivars	320.60 A	191.24 B	75.15 C	36.69 C	32.15 C	24.72 C	198.48 B
LSD values	LSD cultivar* = 11.78		LSD subculture* = 14.08		LSD cultivar x subculture* = 36.70		

Different letters between cultivars denote statistically significant differences (LSD test, p < 0.05)

Axillary shoots formation

There was no statistically significant difference, in terms of average axillary shoots formation, among cultivars regardless of being OP or F₁. On the other hand, there were statistically significant differences among all cultivars regarding axillary shoots formation based on number of subculture (Table 2). Among all evaluated globe artichoke cultivars, 'Imparator' F₁ hybrid had with 12.33 axillary shoots/explant at 7th subculture. Considering the response of OP and F₁ hybrid cultivars to subculture averages, the OP cultivars demonstrated consistent values between each other with respect to axillary shoot formation in different subcultures, while F₁ hybrid cultivars had fluctuations on axillary shoot formation. 'Green Globe' F₁ demonstrated the lowest axillary shoot formation at the 10th subculture due to heavy infection problem.

Table 2. Average axillary shoot formation/explant based on cultivars

No of sub-culture	Cultivars						
	'Bayrampaşa' OP	'Sakız' OP	'Olympus' F ₁	'Madrigal' F ₁	'Sambo' F ₁	'Green Globe' F ₁	'Imparator' F ₁
0	1.00 n	1.00 n	1.00 n	1.00 n	1.00 n	1.00 n	1.00 n
1	2.15 h	2.50 f	1.00 n	1.31 m	1.82 j	1.57 l	2.14 h
2	3.28 d	3.00 e	2.00 i	2.32 g	1.33 m	1.83 j	4.51 wx
3	4.94 t	4.47 xy	4.41 yz	3.37 c	1.66 k	3.72 b	5.26 q
4	4.65 v	5.26 q	5.64 n	5.03 rs	1.97 i	5.93 l	4.37 z
5	4.95 t	7.33 c	7.50 b	4.57 w	4.84 u	7.32 c	4.64 v
6	6.23 i	6.32 h	6.80 f	4.93 t	4.01 a	6.79 f	6.16 j
7	5.36 p	6.16 j	5.83 m	5.36 p	5.67 n	6.27 hi	12.33 a
8	4.94 t	5.68 n	7.13 d	5.67 n	6.74 f	6.64 g	4.98 st
9	5.06 r	6.04 k	6.95 e	6.79 f	5.48 o	5.51 o	4.92 t
10	5.10 r	4.46 xy	4.44 yz	6.28 hi	6.03 k	0 o	4.98 st
Averages of cultivars	4.33	4.74	4.79	4.24	3.68	4.23	5.02
LSD values	LSD cultivar* = 0.26		LSD subculture* = 0.31		LSD cultivar x subculture* = 0.82		

Different letters between cultivars denote statistically significant differences (LSD test, $p < 0.05$)

Each cultivar belonging to OP or F₁ hybrids demonstrated variation in itself. It is clearly demonstrated that applying a certain number of subcultures is a necessity to obtain certain number of *in vitro* plants, regardless of being OP or F₁ hybrid cultivar. Bedini *et al.* (2012) stated that maximum shoot proliferation was obtained after 3rd subculture. Similar reports were also reported by Pacifici *et al.* (2007). Findings of present and previous studies clearly indicated that globe artichoke's proliferative process and number of needed subcultures depends on cultivars.

Leaf formation

When the average number of leaves formed on the explant of the globe artichoke cultivars was evaluated, it was seen that OP cultivars had more positive results; especially 'Sakız' formed the maximum average with 20.8 leaves/explant. When leaf formation considered, the OP cultivars reached to maximum leaf formation/explant at the 5th subculture. It was clearly seen that each F₁ hybrid cultivar predominantly reached to maximum leaf formation/explant at the 4th subculture although two of them reached at 5th and 8th subcultures. According to Pacifici *et al.* (2007), the increase in the number of leaves formed in *in vitro* conditions will cause more water loss and cause difficulties in the adaptation of artichoke plants to *in vivo* conditions. Many previous studies demonstrated that leaves of *in vitro* plants have different morphological structures in a comparison to *in vivo* leaves. Accordingly, since they have different leaf morphology, *in vitro* plants need to adapt themselves to unfavorable environmental conditions of *in vivo* by changes on morphological, anatomical, and physiological features (Brutti *et al.*, 2000; Cavallaro *et al.*, 2007).

Table 3. Average leaf formation/explant based on cultivars

Cultivars							
No of sub-culture	'Bayrampaşa' OP	'Sakız' OP	'Olympus' F ₁	'Madrigal' F ₁	'Sambo' F ₁	'Green Globe' F ₁	'Imparator' F ₁
0	1.00 g	1.00 g	1.00 g	1.00 g	1.00 g	1.00 g	1.00 g
1	3.25 e	16.33 b	3.21 e	2.39 f	3.00 e	3.02 e	4.81 ab
2	5.60 w	11.00 k	9.75 n	5.20 y	3.61 d	6.81 s	5.61 w
3	10.28 lm	20.66 a	11.81 ij	5.04 yza	4.90 zab	5.71 vw	8.82 p
4	10.20 m	16.17 b	15.06 c	7.02 s	4.70 b	5.61 w	11.80 ij
5	12.40 h	20.80 a	11.60 j	6.23 t	4.29 c	9.71 n	6.20 tu
6	11.64 j	13.61 f	11.06 k	6.17 tu	4.32 c	8.21 q	8.15 qr
7	12.00 i	14.79 c	14.02 de	5.92 uv	4.85 zab	7.91 r	8.22 q
8	10.51 l	10.20 m	13.82 ef	5.75 vw	5.26 xy	6.31 t	7.91 r
9	9.19 o	9.80 n	14.17 d	5.67 vw	5.13 yz	5.31 xy	7.90 r
10	9.17 o	13.54 f	13.22 g	5.53 wx	4.90 zab	0 h	8.03 qr
Averages of cultivars	8.66 C	13.44 A	10.79 B	5.08 DE	4.18 E	5.42 DE	7.13 CD
LSD values	LSD cultivar*= 1.13		LSD subculture*= 1.35		LSD cultivar x subculture*= 3.53		

Different letters between cultivars denote statistically significant differences (LSD test, $p < 0.05$)

Evaluation of in vitro rooting, acclimatization and survival percentages of cultivars

In this study, the rooting success of the plantlets after micropropagation stage was also evaluated (Table 4). To achieve well-formed root formation several media combinations were tested. Among rooting media combinations $\frac{1}{2}$ MS medium combined with 10.0 mg L^{-1} IAA + 1.0 g L^{-1} activated charcoal was determined to have positive results in terms of *in vitro* rooting. Although some of the plantlets all of cultivars obtained between 6th and 9th subcultures in the medium used for micropropagation were rooted, their rooting percentages were not enough, so they were not taken into further consideration. Regarding *in vitro* rooting percentage, 'Sakız' OP cultivar was the best, while 'Sambo' F₁ hybrid cultivar had the lowest rooting. At the acclimatization stage it was obvious that F₁ hybrid globe artichoke cultivars adapted to *in vivo* conditions better than OP globe artichoke cultivars. Obtained results clearly revealed that right media combination is a necessity for well root formation and right media combination may vary based on genotype. Similar findings were also reported by Cavallaro *et al.* (2004); Iapichino (2013); López-Pérez and Martínez (2015). As stated in previous studies, the most challenging part of *in vitro* micropropagation studies for globe artichoke is the *in vitro* rooting stage (López-Pérez and Martínez, 2015; Ercan, 2016). As seen in present study, *in vitro* rooting rate is quite low due to genotype-related differences. This is a limiting step for the micropropagation, suggesting that various optimization studies are still needed for *in vitro* rooting (López-Pérez and Martínez, 2015).

In the present study, the acclimatized plant percentages were found to be rather low. In a previous study conducted by Cavallaro *et al.* (2007), it was found that there was a relationship between acclimatization time and acclimatized plants. Researchers revealed that the most favorable percentage of acclimatized plants was obtained in the September plantation time due to thermal conditions. On the other hand, in the current study, the plantation was performed in March. Thus, it is thought that having low level of the acclimatized plant percentages can be attributed to unfavorable thermal conditions in March.

Table 4. *In vitro* rooting, acclimatization and survived plantlets of artichoke cultivars

Cultivars	Percentages of <i>in vitro</i> rooting (%)	Percentages of acclimatization (%)	Percentages of survived plantlets (%)
'Bayrampaşa' OP	5.35 b	81.14 c	8.86 bc
'Sakız' OP	6.47 a	88.80 b	7.54 c
'Olympus' F ₁	1.80 e	88.38 b	10.00 b
'Madrigal' F ₁	4.02 c	100.00 a	10.37 b
'Sambo' F ₁	0.73 f	100.00 a	0 d
'Green Globe' F ₁	2.32 d	100.00 a	0 d
'Imparator' F ₁	2.24 d	73.78 d	12.87 a
LSD values	LSD <i>in vitro</i> rooting* = 2.013; LSD acclimatization* = 17.19; LSD survived plantlets* = 44.81		

Different letters between cultivars denote statistically significant differences (LSD test, $p < 0.05$)

Conclusions

The present study was aimed to comparatively evaluate *in vitro* micropropagation efficiency of 2 important local OP globe artichoke cultivars ('Bayrampaşa', 'Sakız') and 5 F₁ hybrid globe artichoke cultivars ('Olympus', 'Madrigal', 'Sambo', 'Green Globe' and 'Imparator'). The experimental results clearly revealed that OP cultivars and F₁ hybrid cultivars reacted differently to *in vitro* mass micropropagation. Besides genotypic differences, it was also observed the number of subcultures was one of the essential factors that affect *in vitro* multiplication rates in both OP and F₁ hybrids. Although the F₁ hybrid cultivars were ahead of the OP cultivars at the acclimation stage, the multiplication rates and rooting values of the OPs were found almost similar to F₁ hybrid cultivars. Therefore, if there are local OP cultivars well adapted to certain ecological conditions as well as having good yield and fruit quality, they can be used in *in vitro* mass propagation as an alternative to F₁ hybrid cultivars which are expensive and sometimes not easy to find.

Authors' Contributions

Both authors read and approved the final manuscript.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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