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Investigation of appropriate grafting method and plant applications to increase grafting success in cucumber

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Abstract
In grafted seedling production, in addition to the compatibility and performance of the rootstock, the correct selection of the grafting method and the treatments to the rootstocks nd scion are effective on the success of the graft. A three-stage trial was conducted to determine the appropriate grafting method, the effect of root cutting, and some treatments on grafting success in cucumber (*Cucumis sativus* L.). In Experiment I, it was aimed to determine the most appropriate grafting technique for cucumber by using single cotyledon, hole insertion, and tube grafting techniques. The effect of rooted and rootless grafting on grafting success and seedling growth in Experiment II was determined by using the most appropriate grafting technique determined in Experiment I. In experiment III, the effect of sucrose, IBA (Indole-3-butyric acid) and antitranspirant applications on rootstocks on the success of grafting was determined. The graft success rate of the grafted plants was evaluated 14 days after grafting. While the most appropriate grafting technique was the single cotyledon grafting method with a success rate of 76%, the grafting success rate was 67.8% and 55.6% in hole insertion and tube grafting methods, respectively. The effect of grafting with rooted or rootless rootstock on grafting success was found to be insignificant. The highest stem fresh and dry weight were recorded in rooted grafting with 28.00 and 2.30 g/plant, respectively. The highest root fresh and dry weights were found in rooted grafting with 19.30 and 1.93 g/plant, respectively. In Experiment III, the highest grafting success was obtained from sucrose+antitranspirant (98.82%) and sucrose+antitranspirant+IBA (97.65%) applications, respectively. The lowest grafting success was determined in antitranspirant (74.86%) and control (78.24%) applications. According to the results te highest grafting success was achieved by using rooted rootstocks and single cotyledon grafting method. In addition, the combined application of sucrose and antitranspirant and the triple combination of sucrose, antitranspirant, and IBA to rootstocks before grafting is recommended because they increase the success of grafting in cucumber.

Keywords
Cucumis sativus, Rootstock, Auxin, Carbohydrate, Grafting success rate

Introduction
Cucumber (*Cucumis sativus* L.) is an important vegetable belonging to the Cucurbitacea family, which is grown in greenhouses and open fields worldwide. World cucumber production is 91.258.272 tons on an area of 2.261.318 hectares. The cucumber production of Turkey is 1.926.883 tons (FAO, 2020). In greenhouse and open field cucumber cultivation, especially in arid regions, some abiotic stress factors are encountered due to excessive mineral fertilizer application, high pH water use, soil salinity and high temperatures in summer (Fan et al., 2020; Phogat et al., 2020). The negative impact of such stress factors is expected to increase day

by day due to climate change. Abiotic stresses, in general, cause yield losses in agricultural areas of the world and this makes agricultural areas unusable. Up to now in cucumber plants; the effects of many abiotic stresses such as alkaline stress (Nie et al., 2018), drought (Li et al., 2018), salinity (Zhang et al., 2020) and low-high temperature stress (Ali et al., 2018) were examined. However, while these studies focus on a single stress, global climate changes cause multiple stresses in various combinations (Thomas-Barry et al., 2021). Plants are stable organisms and have gradually developed defense systems to protect themselves from environmental stresses (Isah, 2019). In addition, various agricultural practices contribute to adapting to environmental stresses (Singh et al., 2021). One of the approaches used in agriculture to overcome abiotic stresses is grafting on suitable rootstocks (de Oliveira et al., 2021). Inoculation is considered one of the most important cultural techniques that can save time and cost in the fight against abiotic stress factors in breeding programs (Bithell et al., 2013). Vegetable grafting; it is a technique that enables special plant parts (rootstock, scion) to be grown as a single plant by combining them with a suitable technique under appropriate conditions. For this reason, both the scion and rootstock have an effect on the performance of the plant (development, yield, quality, tolerance to stress conditions) in different conditions (Etehadnia et al., 2008). Although grafting is a cultural process that is usually applied to edible vegetables (watermelon, tomato, cucumber, eggplant, melon and pepper), it can also be done in plants whose leaves are consumed recently, such as basil. As rootstocks, genotypes with the desired characteristics within the species can be used, as well as different species and hybrid rootstocks within or between species. Watermelon, melon and cucumber can be grafted onto their wild forms or pumpkin rootstocks (bottle gourd, pumpkin, winter squash, luffa, ficifolia) (Lee, 1994; Yetişir et al., 2004). Inoculation in cucumber plants can increase tolerance to abiotic stress factors, as well as increase the uptake and more effective use of water and nutrients (Sallaku et al., 2019). The choice of grafting method depends significantly on the plant type, the grower's judgment and experience, as well as the facilities available. In the world, generally the methods of approach graft (Sakata et al., 2005), curved cut graft (single cotyledon) (Lee et al., 2010) and punch graft (Lee et al., 2010) are used in cucumber. In Turkey, puncture graft and single cotyledon method are generally used as grafting method in cucumber. The grafting method that is widely used in cucumber seedling plants in our country is the single cotyledon grafting method (Yetisir, 2017). The single cotyledon grafting method is preferred more than other grafting methods because the grafting technique is simple and the grafting can be done faster, and the labor cost is less than other grafting methods. In cucurbits, different grafting techniques are also used, such as using rooted and rootless rootstocks. However, using these grafting techniques (rootless rootstock or rooted rootstock) has several advantages and disadvantages (Lee & Oda, 2002). Rootless grafting method has advantages such as enabling fast and easy grafting for some vegetables, adjusting the rootstock size during grafting for seedling homogeneity and being more hygienic. On the other hand, the disadvantages of this method are; the prolongation of the graft union period, the delay in root formation during the healing of the graft, and the slow vegetative growth in general (Ulaş et al., 2019). Because both graft union and root growth require energy, cutting the roots can allow the energy stored in the rootstock to be used for graft union, resulting in increased graft success (Penny et al., 1976; Lee, 1994; Memmott & Hassell, 2009).

For graft combination, newly grafted plants are kept in low light conditions for at least 3 days, under which conditions the synthesis of new carbohydrates is limited; therefore, the grafted seedling is dependent on stored carbohydrates for survival (Memmott & Hassell, 2009). When both the rootstock and cuttings are cut, the transport of water, phytohormones and carbohydrates stops. The success of grafting depends on the development of vascular tissue (xylem and phloem) and reconnection between rootstock and scion (Pina & Errea, 2005; Aloni et al., 2010; Melnyk, 2017). Carbohydrates in rootstock play a crucial role in callus formation and cellular differentiation that connects vascular bundles at the graft interface (Ogata et al., 2005); According to Rapaka et al. (2007) removing both cotyledon leaves during grafting causes the plant to decrease the amount of stored carbohydrates. The decrease in the amount of carbohydrates reduces the rate of cell division, and as a result, the fusion of tissues is prevented and the graft retention rate decreases (Asahina et al., 2002). On the other hand, since tomato (*Solanum lycopersicum*) and other Solanum species have higher carbohydrate levels in the hypocotyl, the graft retention rate with the tube grafting method is quite high (>95%) (Davis et al., 2008). By applying sucrose solution to rootstock seedlings before grafting, the carbohydrate level in hypocotyl of rootstock could be increased, resulting in an increased percentage of graft retention in watermelon. Dabirian and Miles (2017) reported that sucrose and antitranspirant solution applications to rootstock seedlings before grafting process increased the success of grafting. Abscisic acid (ABA) and antitranspirant products can close the stomata by penetrating the leaf and manipulating the ABA signaling pathway and reduce Scion transpiration (Nitzsche et al., 1991). Reducing scion transpiration during the vascular connection between rootstock and scion during graft fusion is crucial for grafted plant survival (Dabirian & Miles, 2017; Devi et al., 2020). Phytohormones such as auxin and cytokinin are reported to affect the formation and proliferation of callus and new vascular tissues by promoting cell division and/or cell differentiation (Salisbury & Ross, 1992; Preece & Read, 2005). The cotyledons are an important source of auxin, and auxin produced in the cotyledon affects the percentage of graft retention in plants (Bhalerao et al., 2002; Procko et al., 2014; Nanda & Melnyk, 2018). The aim of the study is to determine the effects of three different grafting methods and rooted and rootless grafting, as well as sucrose, auxin and antitranspirant applications to the rootstock before grafting on the success of grafting in cucumber.

Materials and Methods

The study was carried out in the seedling unit of the climate-controlled and fully automated Venlo type glass R&D greenhouse of Kırşehir Ahi Evran University in 2022. Research subjects; grafting method (Experiment I), rooted and rootless grafting (Experiment II), and the effects of different applications on rootstocks on grafting success (Experiment III). The detailed information of the study subjects and the number of plants that were grafted are given in Table 1.

Table 1. Detailed information on the factors used in the experiments and the number of grafted plants.

Experıment	Treatments	Number of grafted plants (number)
Experıment I	Single cotyledon graft	90
	Hole insertion graft	90
	Tube graft	90
Experıment II	Rooted grafting	90
	Rootless grafting	90
Experıment III	Control (Tap water)	60
	Sucrose (S) 2% (w/v)	60
	Antitranspirant (A) 2% (v/v)	60
	Auxin-Indole-3-butyric acid (IBA) 20 mg/l	60
	S+IBA	60
	S+A	60
	A+IBA	60
	S+A+IBA	60

Antitranspirant 2%(V/V): Prohexadione-Calcium 'Velonta' Trade Name (10% Pro-Ca, Basf, Ludwigshafen, Germany), Auxin-Indole-3-Butyric Acid 20 Mg/L (IBA), Waltham, Ma,USA), S+A:(Sucrose 2% (W/V)+Antitranspirant 2% (V/V)), S+IBA:(Sucrose 2% (W/V)+Auxin-Indole-3-Butyric Acid 20 Mg/L), A+IBA:(Antitranspirant 2% (V/V)+Auxin-Indole-3-Butyric Acid 20 Mg/L), S+A+IBA:(Sucrose 2% (W/V)+ Antitranspirant 2% (V/V)+ Auxin-Indole-3-Butyric Acid 20 Mg/L). Sucrose antitranspirant and auxin solutions were applied by dissolving them in tap water.

Plant material and grafting process

The study, Meloni hybrid cucumber cultivar was used as a scion and Kublai hybrid (*C. maxima* x *C. moschata*) was used as rootstock. Scion and rootstock seeds were sown in 45-cell viols, in a mixture of 2:1 peat and perlite (EC: 0.36 dS m-1, pH: 6.0-6.5) with 5-7 days intervals, with the rootstock and the scion being the scion first. Grafting was done when the rootstock and scion reached the stage of 1-2 true leaves.

Grafting methods (Experiment I)

In experiment I, single cotyledon graft, hole insertion graft and tube graft method were used as graft methods. In single cotyledon graft, the growth point of the rootstock, which has reached the grafting stage, was removed and a 8-10 mm long cut with a slight inclination was made at the growth point of the cotyledon leaf from one to the other. The hypocotyl of the scion was cut with the same slope and the cutting surfaces were attached with the help of a grafting forceps and the grafting process was completed (Oda et al., 1993; Davis et al., 2008).

Hole insertion graft, the first true leaf and growth point of the rootstock were removed, a 1.4 mm diameter metal hole was drilled with a pointed tip, and both sides of the hypoquitila were cut at an angle of 35°-45° 1-2 cm below the cotyledon leaves. It was built by placing it (Yetisir, 2001). The tube graft was cut inclined from under the cotyledon leaves of the rootstock at the 1-2 leaf stage, and then the graft tube was placed on the cut rootstock, half empty from above. The scion, which was cut at the same angle from about 1 cm below the cotyledon leaves, was made by placing it on the empty part of the tube in such a way that the cutting surfaces were completely in contact (Figure 1).

Rooted and rootless grafting (Experiment II)

In experiment II, the rooted and rootless grafting method was used, according to the results of experiment I, the single cotyledon graft method, which had the highest success rate of grafting. In rooted grafting, grafting was done on the viol where the rootstocks were. In rootless grafting, rootstock roots were cut from the point where the hypocotyl ended and replanted in viols containing a 2:1 mixture of peat and perlite after the grafting process was completed (Figure 1).

Sucrose, Antitranspirant and IBA application (Experiment III)

In Experiment III, single choyledon and rooted grafting methods were used according to the results of Experiments I and II. Experiment III, sucrose, antitranspirant and Auxin-IBA were applied before grafting on rootstock seedlings. 6 days before grafting on rootstock seedlings, 50 ml of sucrose (2%) was applied per plant. The sucrose application was diluted with water containing sucrose (2%) in 3 times, 20 ml 6 days before the grafting, 20 ml 4 days before and 10 ml 2 days before the grafting. Antitranspirant (2%), which slows down the plant transpiration rate, was used at 50 ml per plant before grafting on rootstock seedlings. Antitranspirant was given by spraying on rootstock leaves in 20 ml 6 days before grafting, 20 ml 4 days before and 10 ml 2 days before grafting. Auxin (IBA) 10 mg/l was used. Auxin application was given by spraying 4 ml 6 days before grafting, 4 ml 4 days before and 2 ml 2 days before grafting. The leaves of the control plants were sprayed with the same amount of distilled water.

Figure 1. Grafting techniques, a) single cotyledon grafting, b) hole insertion grafting, c) tube grafting, and d) rootless grafted seedling

After all grafting processes completed, the grafted plants were placed in the post-grafting care unit, which was shaded with 90-95% relative humidity, 22-25 ºC temperature and 50% light transmission shade material by spraying water on the grafted plants. three days after grafting, the shade material and plastic inoculum containers were opened in the morning and evening, this exercise was done for 7-10 days, and on the 10th day the plants were removed from the grafting unit.

Plant Measurements and Data Analysis

Determination of grafting success rate (%) values

The graft retention rate of the grafted plants was evaluated 14 days after grafting. Scion and rootstock leaves erect or parallel to the stem and robust marketable plants were considered viable. Plants with severely wilted scion and rootstock were considered dead. Grafting success rate was calculated with the following formula = (Number of live plants/total number of grafted plants) × 100%.

Researching of hypocotyl, Cotyledon, First true leaf, stem, root, fresh and dry weight characteristics

Researching of hypocotyl, cotyledon, first true leaf, stem, root fresh and dry weights features was carried out in three replications and 20 plants in each replication, in a total of 60 plants.

Hypocotyl length (mm)

The distance between the soil surface and the horizontal cotyledon leaves was measured with a digital caliper and recorded as the length of the hypocotyl when the zucchini seedlings were at the grafting stage.

Hypocotyl thickness (mm)

The cotyledon, which is the graft junction, was measured with a digital caliper just below the leaves and parallel to the leaves and recorded as hypocotyl thickness.

Cotyledon length (mm)

The distance from the junction of the cotyledon leaves with the plant body to the tip of the leaf was measured with a digital caliper and recorded as the cotyledon length.

Cotyledon width (mm)

The leaf width at the midpoint of the cotyledon leaves was measured with a digital caliper and recorded as the cotyledon diameter.

First true leaf width (mm)

Just before the first true leaf grafting, the width in the middle part of the leaf was measured with a digital caliper.

Root and stem fresh and dry weights (g)

14 days after grafting, the grafted plants were separated into their organs as roots and stems, their fresh weight was measured with the help of a digital scale Dry weights were determined after three days in an oven at 65°C.

Data Analysis

The data obtained in the research were subjected to variance analysis according to the randomized plot design with the IBM SPSS statistical program, and the parameters with significant F values were grouped according to Duncan test at 5% and 1% significance level. The averages shown with the same letter in the tables are statistically in the same group. Correlation analysis was performed using SPPS computer package statistical program on the data obtained from seedling characteristics and grafting success measurements (SPSS, 2013).

Results and Discussion

In the study carried out to determine the most suitable grafting technique for cucumber (Experiment I), single cotyledon, hole insertion, and tube grafting techniques were compared. For each grafting method, totally 90 plants were grafted. The grafting was successful in 69 plants in the single cotyledon grafting method, 50 plants in the tube grafting method and 61 plants in the perforated grafting method. The highest grafting success rate was 76.7% in the single cotyledon grafting method, followed by the hole insertion graft method with 67.8%. The lowest grafting success was determined in the tube grafting method with 55.6% (Table 2). Since the cost of grafted cucumber seedlings is higher than that of ungrafted seedlings, the survival rate after grafting should be high. The grafting techniques used, rootstock/scion combination, pre- and post-grafting care conditions affect the success of the graft in cucumber. Single cotyledon grafting method is preferred in cases where rootstocks have thin stems, especially watermelon, cucumber and melon (Sakata et al., 2007). This method is done when the rootstock and scion are of similar size and the first true leaf of the rootstock has started to develop (Oda, 1999; Lee & Oda, 2002). Most cucurbits can be grafted onto several rootstocks, and incompatibility can be corrected using appropriate grafting methods and growing media. Rojas and Riveros (1999) reported that the grafting method and the variety used significantly affect the success of the graft. Oda et al. (2001) reported that grafting methods in cucumber, the number of cotyledons left on the rootstock, the width of the graft surface cutting area and the compatibility of the diameters of the rootstock and scion affect the success of the graft. Oda et al. (2001) reported that grafting methods in cucumber, the number of cotyledons left on the rootstock, the width of the graft surface cutting area and the compatibility of the diameters of the rootstock and scion affect the success of the graft.

Table 2. Experiment I, grafting method, Number of grafted plants (number), Grafting success and rate

Grafting Method	Number of grafted plants (number)	Grafting success (number)	Grafting success rate (%)
Single cotyledon graft	90	69	76.70a
Tube graft	90	50	55.60c
Hole insertion graft	90	61	67.80b
F values:			**

F values: $p < 0.01$ (**) and N.S. Non Significant

According to experiment I results, it was determined that the most appropriate grafting method in cucumber was the single cotyledon grafting method. In experiment II, the plants were grafted with and without roots, with 90 plants in each, using the single cotyledon grafting method. While grafting of 69 plants was successful in rooted grafting, grafting of 70 plants was successful in rootless grafting. However, the effect of rooted and rootless grafting on the success of the grafting was not found to be statistically significant. When the effect of rooted and unrooted grafting on stem fresh and dry weight was examined, the highest stem fresh weight (28.00 g) and stem dry weight (2.30 g) were obtained in rooted grafting. In rootless grafting, the fresh weight of the stem was 16.30 g and the dry weight of the stem was 1.33 g. When root fresh and dry weights were examined, the highest values were obtained in root grafting in the same way. In rooted grafting, root fresh weight was measured as 19.30 g and root dry weight was measured as 1.93 g. In rootless grafting, these values were determined as 13.00 g and 1.33 g, respectively (Table 3). Rootless grafting increases the amount of primary roots and as a result, the plant develops vigorously as it increases the plant's tolerance to cold and heat (Lee & Oda, 2002). Today, more than 40% of watermelon grafting is done by this method (without roots) in Japan (Devi et al., 2021). The survival rate in rootless grafted plants is 11% lower than in rooted grafting and rootless grafting has advantages such as preventing the risk of contamination with grafting machines and rapid grafting (Devi et al., 2021). Ulaş et al. (2019) suggested that grafting on rootless rootstocks for growth and physiology of watermelon plants grown under hydroponics conditions is not a useful implementation strategy, even if they are grafted on strong rootstocks. In our study, it was observed that rootless grafting of cucumber did not have a negative effect on the success of the graft. For this reason, rootless grafting method can be recommended as it will be beneficial in preventing the risk of contamination in grafting machine/robot applications and will reduce the aged/rolled root ratio.

Table 3. Experiment II, Number of rooted and rootless grafted plants, Grafting success and rate, Stem fresh weight, Stem dry weight, Root fresh weight and Root dry weight.

Rooted, Rootless Grafting	Number of grafted plants (number)	Grafting success (Number)	Grafting success rate (%)	Stem fresh weight (g/plant)	Stem dry weight(g)	Root fresh weight (g)	Root dry weight (g)
Rooted graft	90	69	76.70	28.00a	2.30a	19.30a	1.93a
Rootless graft	90	70	77.70	16.30b	1.33b	13.00b	1.33b
F values:			N.S	**	**	**	**

F values: $p < 0.01$ (**) and N.S. Non Significant

Considering the effects of the applications on the success of grafting in experiment III, the highest graft success was obtained from sucrose+antitranspirant (98.82%) and sucrose+antitranspirant+IBA (97.65%) applications, respectively. The lowest grafting retention rate was obtained from antitranspirant (74.86%) and control (78.24%) applications (Figure 2). In the effect of applications on hypocotyl thickness, the highest hypocotyl thickness was determined in IBA application alone (4.61 mm), while the lowest hypocotyl thickness was determined in plants with sucrose application alone (3.66 mm) and in rootstocks that were not treated (3.69

mm). The highest hypocotyl length was measured in untreated (control) rootstocks (40.91 mm) and antitranspirant +IBA (40.81 mm) application, while the lowest was measured in antitranspirant alone (31.89 mm) application. The highest cotyledon width was obtained in sucrose+antitranspirant (43.69 mm), antitranspirant+IBA (42.52 mm) and sucrose+antitranspirant +IBA (40.56 mm) applications. The highest cotyledon length was obtained from antitranspirant alone (58.32 mm) and antitranspirant+IBA (57.58 mm) applications. The first true leaf width was highest in IBA alone (80.05 mm) application, while the lowest first true leaf width was determined in sucrose+antitranspirant (40.78 mm) application (Table 3).

Karaağaç (2013) reported that the short hypocotyl length is due to the genetic structure of the plants. Hypocotyl length is less important than hypocotyl thickness in terms of grafting success. However, the hypocotyl lenght should not be too short for easier grafting. In addition, there are disadvantages such as slipping in the grafting site and lying on its side, since the grafting apparatus will be attached higher in rootstocks with long hypocotyls (Oda, 1994; Hamamoto & Oda, 1997; Yang et al., 2012; Yıldız & Balkaya, 2016). Hypocotyl lengths can be controlled by applying different chemicals and regulating environmental conditions (Yang et al. 2012). Many researchers reported that the difference between the hypocotyl thickness and the number of vascular bundles among rootstock and scion did not have a significant effect on graft success (Edelstein et al., 2004; Yetişir & Sarı, 2004; Yetişir et al., 2007). Hypocotyl thickness at the time of graft; it may vary depending on the grafting time of the rootstock, the grafting method, the light and temperature values of the environment where the seedlings are grown (Yıldız & Balkaya, 2016). Since the first true leaf is removed in the single cotyledon grafting method, the carbohydrate reserve of the first true leaf cannot be utilized. Therefore, the over development of the true leaves of the rootstock seedlings before grafting may adversely affect the success of the graft. By applying sucrose solution to rootstock seedlings before grafting, the carbohydrate level in rootstock hypocotyl can also be increased, resulting in increased success of watermelon grafting (Dabirian & Miles, 2017). According to Devi et al. (2020) reported that the grafting success of plants applied sucrose and antitranspirant solutions before grafting to rootstock seedlings was 91%, 2% sucrose application alone was 67%, and plants irrigated with only water were 25%. Researchers also reported that the success of the graft was 70% when they applied a different sucrose-free antitranspirant solution to the plants. Antitranspirant products containing abscisic acid (ABA) enter the leaf, manipulate the ABA signaling pathway and cause stomatal closure. As a result, scion-rootstock transpiration is reduced (Nitzsche et al., 1991; Grill & Ziegler, 1998; Hetherington, 1998). In addition to carbohydrates and ABA, other phytohormones play an important role in graft success (Kümpers & Bishopp, 2015; Melnyk, 2017). Previous studies have shown that phytohormones such as auxin and cytokinin stimulate cell division and/or cell differentiation, triggering the proliferation of callus and new vascular tissues (Salisbury & Ross, 1992; Preece & Read, 2005). The cotyledons are an important source of auxin, and auxin derived from the cotyledon promotes graft union in young plants (Bhalerao et al., 2002; Nanda & Melnyk, 2018). Since scar tissue is opened in plants during grafting, external auxin application to the plant increases the rate of cell division and supports graft union (Procko et al., 2014).

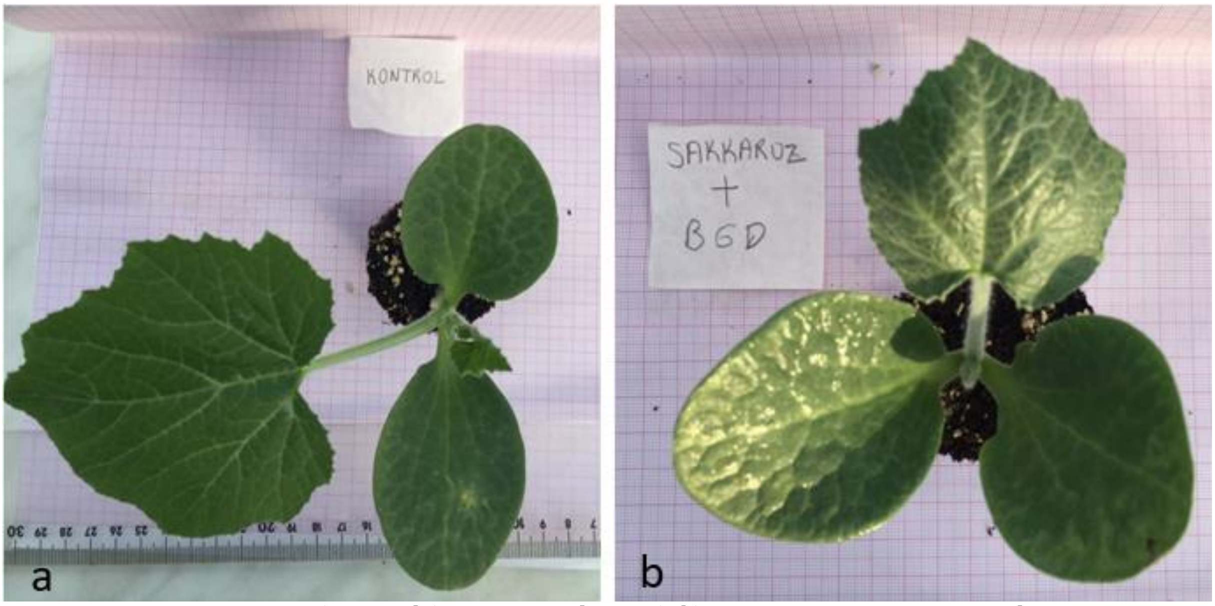

Figure 2. a) Control (tap water application), b) Sucrose+Antitranspirant application

In the study, the highest graft success was obtained in sucrose + antitranspirant and sucrose + antitranspirant + IBA applications. The reason for this is that the cotyledon widths of the rootstock with sucrose and antitranspirant application are higher than the other applications and the first true leaves are small. The width and length of the cotyledon leaves increased the success of graft union, since cotyledons are the carbohydrate storage of the plant and grafting was done using a single cotyledon in the single cotyledon grafting method.

Table 4. Experiment III, Effects of treatments on hypocotyl thickness, hypocotyl length, cotyledon width, cotyledon length, first true leaf width and graft success rate (%)

Treatment	Hypocotyl thickness (mm)	Hypocotyl length (mm)	Cotyledon width (mm)	Cotyledon length (mm)	First true leaf width (mm)	Graft success rate (%)
Rootstocks Kublai Control(Tap Water)	3.69d	40.91a	39.65cd	54.60	67.60b	78.24e
Sucrose	3.66d	35.70ac	41.84a-c	50.29	52.88c	85.29d
IBA	4.61a	41.25a	38.37d	53.54	80.05a	85.88d
Antitranspirant	3.87bcd	31,89c	42,79bd	58.32	65.81b	74.86f
S+IBA	3.67d	38,40ab	40,63a-d	51.06	49.12cd	91,43c
S+A	3.73cd	32.99bc	43.69a	55.77	40.78d	98.82a
A +IBA	4.21b	40.81a	42.52ab	57.58	65.04b	93.71b
S+IBA+A	4.07bc	38.49ab	40.56a	54.42	44.52cd	97.65a
F values	**	**	**	N.S.	**	**

F values: $p < 0.05$ (*), $p < 0.01$ (**) and N.S. Non Significant

The relationships between the measured seedling characteristics and the graft success rate were evaluated in terms of statistical significance based on the error margin of P≤0.05 and P≤0.01 (Table 5). A significant positive correlation was determined between hypocotyl thickness and cotyledon length, true leaf width, and graft success rate (correlation coefficients 0.325*, 0.447**, 0.298*, respectively). There was a significant but negative correlation between hypocotyl length and cotyledon width, cotyledon length and graft success rate (correlation coefficients were 0.547**, 0.403**, 0.840**, respectively). A significant positive correlation was found between cotyledon width and cotyledon length and graft success rate (correlation coefficients; 0.456**, 0.889**, respectively). The correlation value between cotyledon length and graft success rate is 0.335*. While a significant positive correlation was observed between the graft success rate and the hypocotyl thickness, cotyledon width and cotyledon length values (correlation coefficients were respectively; 0.298*, 0.889**, 0.335*), the same feature; showed a significant but negative correlation with hypocotyl length (correlation coefficient; 0.840**).

Table 5. Correlation coefficients between seedling characteristics and graft success rate parameters

	Pamaterers	1	2	3	4	5	6
1	Hypocotyl thickness	1	0,169	0,251	0,325*	0,447**	0,298*
2	Hypocotyl length		1	-0,847**	-0,403**	-0,007	-0,840**
3	Cotyledon width			1	0,456**	-0,199	0,889**
4	Cotyledon length				1	0,02	0,335*
5	First true leaf width					1	-0,109
6	Graft success rate						1

F values: $p < 0.05$ (*), $p < 0.01$ (**) Significant

Conclusion

Since the cost of grafted cucumber seedlings is much higher than that of ungrafted seedlings, the survival rate after grafting should be high. The low graft success rate is the biggest barrier to the use of grafted plants in most countries as it exacerbates the high cost. The grafting techniques used, rootstock/scion combination, pre- and post-grafting care conditions affect the success of the graft in cucumber. According to our study results, the highest graft success in cucumber; it was obtained in rooted grafting method with single cotyledon graft and sucrose+antitranspirant and sucrose + antitranspirant + IBA applications to rootstocks before grafting. In the future, studies should be carried out to increase the carbohydrate reserves in the hypocotyl and cotyledon of cucurbit rootstocks, as well as to improve the optimum environmental conditions such as temperature, relative humidity and light in the post-grafting care room and to increase the rooting rate of rootless grafted plants.

Compliance with Ethical Standards

Conflict of interest

The authors declared that for this research article, they have no actual, potential or perceived conflict of interest.

Author contribution

The contribution of the authors to the present study is equal.

All the authors read and approved the final manuscript. All the authors verify that the text, figures, and tables are original, and they have not been published before.

Ethical approval

Ethics committee approval is not required.

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Not applicable.

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Not applicable.

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