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Research Article Dergimiz yazarlarından Ramazan Şadet GÜVERCİN'in vefat haberini almış bulunmaktayız. Merhuma Allah'tan rahmet, sevdiklerine başsağlığı diliyoruz.

Vegetative Propagation of Cotton (Gossypium spp.) By Rooting

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Keywords Clone production, Cotton, Hybrid vigor, Rooting **Abstract:** Cotton plant is produced with seeds. However, its genetic structure is suitable for vegetative production. In this study, rooting method used to propagate cotton plant as vegetative. Experiment was conducted in climate chamber at East Mediterranean Transitional Zone Agricultural Research Institute in 2020. Stems of Askabat 100 (*G. barbadense* L.), Stoneville 468 (*G. hirsutum* L.) cultivars and their F₁ hybrid were used as plant materials while 2000 ppm IBA (Indole butyric acid) concentration used as chemical materials. As a result of the study, it was determined that cotton genotypes rooted and formed shoots in different rates. Moreover, F₁ hybrid produced more clones than their parents, followed by Askabat 100 and Stoneville 468. Also, F₁ hybrid were showed that superior heterotic effects as heterosis and heterobeltiosis in term of investigated properties. This result showed that cotton cultivars and hybrids can be propagation as clone using by rooting, and these clones could be used as rootstocks again in clone production.

Pamuk (Gossypium spp.) Bitkisinin Köklendirme Yöntemiyle Vejetatif Çoğaltılması

Anahtar Kelimeler Klon üretimi, Pamuk, Hibrit gücü, Köklendirme **Öz:** Pamuk bitkisi tohumlarla üretilir. Ancak genetik yapısı vejetatif üretime uygundur. Bu çalışmada, pamuk bitkisinin vejetatif olarak çoğaltılması amacıyla köklendirme yöntemi kullanılmıştır. Deneme, 2020 yılında Doğu Akdeniz Geçit Kuşağı Tarımsal Araştırma Enstitüsü Müdürlüğü'nün iklim odasında yürütülmüştür. Bitki materyali olarak Askabat 100 (*G. barbadense* L.), Stoneville 468 (*G. hirsutum* L.) çeşitleri ile bunların F₁ hibritlerine ait gövde parçaları, kimyasal material olarak ise 2000 ppm IBA (Indol butirik asit) konsantrasyonu kullanılmıştır. Çalışma sonucunda pamuk genotiplerinin sürgün oluşturduğu ancak farklı oranlarda köklendiği belirlenmiştir. Ayrıca F₁ melezi, ebeveynlerinden daha fazla kök üretmiş, bunu Askabat 100 ve Stoneville 468 çeşitleri izlemiştir. Dahası F₁ melezleri incelenen özellikler yönünden önemli heterotik etki (heterosis ve heterobeltiosis) göstermiştir. Bu sonuç, pamuk çeşitlerinin ve hibritlerinin köklendirme yoluyla klon olarak çoğaltılabileceğini ve bu klonların klon üretiminde tekrar anaç olarak kullanılabileceğini göstermiştir.

1. INTRODUCTION

The Gossypium L. genus contains more than 50 species in the world. One of them is G. hirsutum L. (Upland cotton) which different varieties of it were cultivated in Turkey. Turkey is major G. hirsutum L. producers with Greece and Spain in Europe. Moreover, Bulgaria has been producing some amount of cotton. Farming of cotton starts with its seed planting, and quality seeds are bases of modern farming of cotton. But, preparing of quality seeds starts with breeder seeds, and takes minimum five years to serve for farmers with systematic and its expensive methods after breeding studies that takes 8-12 years. Cotton is having sexual reproduction system, but it can rapidly propagation by vegetative methods too such as T or Shield budding method [1]. However, because of easy sowing of cotton seeds with machine, in traditional, grafting, transplantation and pruning practices are not widely used in cotton farming. Whereas, vegetative propagation of cotton is get some advantages. For example, because of every new individual (clone) has the same genetic constitute with its parent, breeders will can save their important hybrids or genotypes along with years in the greenhouse. Moreover, cotton farming will can be done with clones some areas having problems such as salty, droughty and contaminated with Verticillium dahliae Kleb. In this areas, if varieties have strong root development, tolerant to disease with high yield and fiber quality, can be propagated with rooting, furthermore because have strong root system these cultivars can be used as rootstock plant in grafting methods.

On the other hand, for production of cotton seed, rooting, grafting or combination of rooting with grafting can be used in greenhouse. This production is very important. When breeders use this method, will can offer seeds having same genetic potential which produced from lots of clone plants to farmers. So, farmers will know about yield and quality potential of their field before harvest. There are a lot of studies on propagation of cotton by vegetative. Numerous cotton clones have been produced via micro-propagation from shoot [2], cotyledon explants [3] and hypocotyl of cotton seedlings [4]. Moreover, cotton was propagation as vegetative by grafting method in Turkey [1, 5]. The aim of this work is to search possibility of vegetative propagation on cotton (Gossypium spp) by rooting.

2. MATERIALS AND METHODS

2.1. Plant Materials

Stoneville 468 (G. hirsutum L.), Askabat 100 (G. barbadense L.) and their F1 hybrid were used as plant material in this experiment. Askabat 100 needs longer vegetation period for productions cotton, but produced lower seed cotton yield, lint yield and ginning out-turn in Turkey. On the other hand, Stoneville 468 is an upland cultivar having extra seed cotton yield; lint yield and ginning out-turn than Askabat 100, but lower fiber qualities.

2.2. Field Experiment and Climate Chamber Applications

This experiment was conducted at East Mediterranean Transitional Zone Agricultural Research Institute in 2020 (370 38' N; 360 37' E, altitude: 568 m), in climate chamber between 15 October 2020 and 30 November 2020, according to Randomized Complete Block Design (RCBD) with three replications. Plant stems of 22 weeks were used in study. The stems are divided into pieces with at least 4 nodes. Genotypes were represented by 100 plant stem parts in replications. Plant pieces were kept in indole butyric acid (2000 ppm) for 5 minutes and then transferred to perlite medium prepared before in the climate chamber. Then the climatic room conditions were fixed at 28+/-2 oC temperature and 70-80% humidity. With the counts made on 30 November 2020, the number of rooting plants, root number, root length, shoot number and shoot length were determined. Data were analyzed for number of rooting plant, root number per plant, root length per plant (cm), shoot number per plant, shoot length per plant (cm). Then, means of investigated properties were compared by Least Significant Degree at 0.05 (LSD0.05). In addition, hybrid vigor (heterosis and heterobeltiosis) values were determined by the following Eq.1 and Eq.2 formulas at below.

3. RESULTS

3.1. Rooting Stages

In the study, the visuals about rooting can be seen in Figures 1, 2, 3, 4, 5 and 6. Plant parts with or without wood (bark) texture developed callus and rooted (Figure 1 and Figure 2).



Figure 1. Callus formation



Figure 2. Rooting from callus

Thick plant parts formed mostly roots and long shoot (Figures 3 and 4). IBA 2000 dose and duration were sufficient for callus formation (Figures 1 and Figure 2). Furthermore, plant parts forming roots and shoots adapted to pots filled with 1/2 peat + 1/2 soil (Figure 6).



Figure 3. Rooting from green and non-green cuttings



Figure 4. Advanced roots



Figure 5. Clone plant with developed roots and shoots



Figure 6. Clone plant in pots

3.2. Number of Rooting Plant

The variance analysis regarding the number of rooting plants is shown from According to Table 1, the number of rooting plants differed according to their genotypes. The F1 hybrid was the most rooted genotype with 54.88%, and it was followed by Askabat 100 with 49.22% and Stoneville 468 with 26.88% (Table 2). According to Ouma et al. [4], cotyledon node explants of Stoneville 474 have rooted on sucrose based medium + IBA and +/- activated charcoal after a 25, 39 and 65 cumulative days in culture. It is a plant that can reproduce by cotton grafting and micro propagation methods. This study showed that the stem of cotton plant can be rooted and reproduced vegetative. This finding is very important. Wood texture or green plant body parts did not differ and were rooted.

The method will help breeders easily reproduce the plants they want to use for many years in seed production. In addition, they will be able to reproduce and protect important breeding materials in a short time. Moreover, seeds of commercial varieties will be propagated from clones of a plant and given to farmers. Thus, efficiency and quality losses will be reduced. On the other hand, triploid genotypes developed by interspecies hybridization will be easily reproduced by the method and can be used for many years in polyploidy studies.

3.3. Root Number Per Plant

Differences of cultivars were found important for average root number per plant (Table 1). The root number per plant has changed from 19.22 (Stoneville 468) to 37.56 (F1 hybrid).

-	Sources	Degree of freedom	Number of rooting plant	Root number per plant (cm)	Root length per plant	Shoot number per plant	Shoot length per plant (cm)	
	Replication	2	93.78	14.77	1.77	1.33 *	0.45	
	Cultivars	2	657.44 *	253.45 *	40.78 *	1.33 *	70.78 *	
	Error	4	23.61	18.44	3.11	0.17	3.61	
-	Total	8	199.61	76.28	12.19	0.75	19.61	

Table 1 Made 1 . . .

Table 2. Values of inve	stigated properties in ter	rm of rooting applicat	tions						
Cultivars	Number of rooting plant (number)	Number of root per plant (number)		Root length per plant (cm)		Shoot number per plant		Shoot length per plant (cm)	
Stoneville 468	26.88 b	19.22	b	9.89	b	4.00	b	7.11 b	
Askabat 100	49.22 a	27.22	b	12.89	b	4.66	b	11.10 b	
F1 hybrid	54.88 a	37.56	а	17.22	a	5.33	a	16.77 a	
_CV (%)	9.70	15.65		12.80		10.20		15.98	
LSD _{0.05}	11.01	9.74		3.99		0.93		1.53	

CV (%):Coefficient of variation, LSD: Least significant degree



Figure 7. Hybrid vigor (Heterosis and heterobeltiosis) values of F1 hybrid

Also, Askabat 100 has produced 12.89 roots per plant. The F1 hybrid has most number of roots than Askabat 100 and Stoneville 468 having minimum numbers of root (Table 2). Cotton has a rapidly growing taproot and the seedling can reach 20-25 cm deep without even growing above the ground. The final depth of the root system depends on soil moisture, aeration, temperature and variety, but is usually about 180-200 cm. Although cotton stem pieces are rooted, they cannot taproot form like a normal cotton plant.

3.4. Root Length in Plant

Differences of mean of root length of cultivars were found very important (Table 1). According to Table 2, means of root length of cultivars were differed. F1 hybrid has development very long roots than Stoneville 468 and Askabat 100. While F1 hybrids getting 17.22 cm root length, the Askabat 100 and Stoneville 468 cultivars had 12.89 cm and 9.89 cm root length, respectively. Root length is very important to reach water and nutrients in the soil. Furthermore, the number of roots is also important in terms of the plants holding in the soil and resisting wind effects.

3.5. Shoot Number Per Plant

In the study, where the average number of shoots varied between 4.0 and 5.33 (Table 2), and difference among genotypes was found to be significant (Table 1). While the F1 hybrid is produce 5.33 shoot, Askabat 100 and Stoneville 468 have produced 4.66 and 4.00 shoots,

respectively. The high numbers of shoots were found important in terms of photosynthesis. Özyiğit [2] reported that shoot development was maximum 74.2 % in cotyledon nodes.

3.6. Shoot Length per Plant

Plant shoot lengths of genotypes were found to be significant (Table 1). The F1 hybrid had the longest shoots, followed by Askabat 100 and Stoneville 468. This situation may have occurred due to genetic difference. Shoot length of F1 cross was 16.77 cm, while shoot length of Askabat 100 and Stoneville 468 were 11.10 cm and 7.11 cm, respectively. Plant shoot length was found to be related to shoot number. Those with more plant shoots formed shorter shoots.

3.7. Hybrid Vigor (Heterosis and Heterobeltiosis)

The hybrid vigor of the F1 hybrid for the traits examined is monitored from Figure 7. According to Figure 7, high heterosis and heterobeltiosis values were found to be significant. The highest heterosis and heterobeltiosis were observed in plant shoot length. It was followed by plant root number and plant root length. The importance of hybrid power of root number and length was found to be important in terms of clone production in cotton plant. The F1 hybrid produced 50% more shoots, 38.75% more root count, 32.50% more root length and 10.18% more clones than its superior parent.

4. CONCLUSIONS

Cotton plant is produced with seeds. Lots of studies have shown that cotton can also be produced by vegetative propagation methods. While cotton is propagated by grafting method [1, 6-7], Özyiğit [2] produced vegetative from internodes under in vitro conditions. In addition, Karaca et al. [5] had been successful in seedling with grafting. In addition, Karaca et al. [5] had been successful in seedling with grafting. In this study, another vegetative reproduction method has been experiment, which rooting. At the end of study, it was determined that not only cultivars belonging to G. hirsutum L but also G. barbadense L with their F1 hybrid had been produced by rooting. Moreover, F1 hybrid was found more important than their parents in tern of investigated properties, showed more important heterotic effects (heterosis and heterobeltiosis). This may be due to heterozygous genetic makeup of the F1 hybrid. The results of study showed that rooting can be done on cotton in advanced plant form. On the other hand, the fact that F1 hybrid is superior to its parents in terms of root number and root length has shown that F1 crosses can be used as rootstock for clone production. Thus, quality seeds will be provided to farmers by producing clones of productive and high quality varieties on clone rootstock.

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