Rootstock Resistance to Fusarium Wilt and Effect on Watermelon Fruit Yield and Quality

Halit Yetışır,[1] Nebahat Sari[2] and Seral Yücel[3]

The potential of grafted watermelon for resistance to *Fusarium oxysporum* f.sp. *niveum* on some Curcurbitaceae, *Lagenaria, Luffa, Benincasa* and commercial rootstocks was evaluated. Effects of grafting on yield and quality of diseased plants were evaluated. All grafted plants and rootstocks were resistant to the three known races (0, 1, and 2) of *F. oxysporum* f.sp. *niveum* except watermelon cv. 'Crimson Tide', which was susceptible to race 2. Fruit yield was positively (21–112%) affected by *Lagenaria* rootstocks but negatively affected (200–267%) by *Cucurbita* rootstocks when compared with the control. While only minor differences in fruit quality were determined in control and grafted plants on *Lagenaria* rootstocks, the quality parameters for watermelon grafted onto *Cucurbita* rootstocks were lower than in the control. The reasons for low yield and quality might be due to an incompatibility between *Cucurbita* rootstocks and watermelon. These results showed that rootstock influence on disease resistance as well as yield and quality of scion fruit is important in determining the potential use of grafting applications in watermelon.

KEY WORDS: Grafting; *Fusarium oxysporum* f.sp. *niveum*; watermelon; disease resistance; yield and quality.

INTRODUCTION

Watermelon (*Citrullus lanatus* (Thunb.) Matsum. and Nakai) is one of the most widely cultivated crops in Turkey. Turkey is the second largest watermelon-producing country in the world (after China), with a production rate of 4 million tons per year (1). Soilborne diseases such as Fusarium wilt caused by *Fusarium oxysporum* f.sp. *niveum (Fon)* may have strong adverse effects on watermelon production either during early cultivation under plastic tunnels or later in the season under open field production conditions because of continuous and intensive cropping (11,22). Three races (0, 1, and 2) of *Fon* have been identified in the Çukurova region, where 35% of the watermelon production in Turkey occurs (22). Resistant watermelon cultivars could be introduced from combined breeding programs designed to prevent the occurrence of soilborne diseases (15). However, developing new cultivars resistant to diseases is time consuming and enhances the danger of the resistant cultivars becoming susceptible to new races of pathogens. On the other hand, grafting susceptible cultivars onto resistant rootstocks may enable the control of soilborne diseases (13,18). Grafting was first performed in vegetable plants in Japan and Korea in the

Received Aug. 21, 2002; received in final form Nov. 27, 2002; http://www.phytoparasitica.org posting Feb. 2, 2003.

[1]Dept. of Horticulture, Faculty of Agriculture, University of Mustafa Kemal, Hatay, Turkey. *Corresponding author [Fax: +90-326-245-58-36; e-mail: yetisir1@yahoo.com].

[2]Dept. of Horticulture, Faculty of Agriculture, University of Çukurova, Adana, Turkey [e-mail: nesari@mail.cu.edu.tr].

[3]Adana Plant Protection and Research Institute, Adana, Turkey.

late 1920's by grafting watermelon onto gourd rootstocks (13). In the last 20 years, grafted plant cultivation has largely increased for both Solanaceace and Cucurbitaceae, especially when adequate disease resistance is not available in commercial hybrids (13). Cultivation of grafted vegetables has been done mostly in Korea, Japan and some Asian and European countries where land use is very intensive and continuous cropping is common (13,18). After methyl bromide was banned in many countries, the use of grafted seedlings was introduced in Turkey. Seedling companies started to produce tomato and cucumber grafted seedlings. Research has continued on watermelon, melon and cucumber grafting. It is believed that use of grafted plants in fruit-bearing vegetables may help to reduce the need for soil fumigation with methyl bromide for many crops.

Plant growth, yield and fruit quality can also be influenced by rootstock. Both fruit size and harvest duration of watermelon cultivars grafted onto rootstocks with vigorous root systems were significantly increased compared to those of ungrafted plants. The influence of rootstocks on fruit quality can often be detrimental, but the results vary greatly, depending on the scion cultivars (13).

In this study, *Fon* resistance of grafted watermelon on Cucurbitaceae, Lagenariae, *Luffa* and *Benincasa* species along with some commercial hybrid rootstocks was determined. Effects of the selected genotypes were evaluated for productivity and fruit quality of watermelon.

MATERIALS AND METHODS

Plant materials Watermelon [*Citrullus lanatus* (Thunb.) Matsum. and Nakai] cv. 'Crimson Tide' susceptible to *Fon* was grafted onto 11 different rootstocks. Name, definition and source of rootstocks are given in Table 1. Ungrafted Crimson Tide was used as the control. The hole insertion grafting technique (peg) was used and plants were grafted by following the procedure described by Lee (13).

Inoculation tests Artificial inoculation was performed under controlled conditions (24±1°C, 16 h light/ 8 h dark photoperiod) on 15 seedlings of each genotype (rootstocks and scion) at the first true leaf stage. The inoculum used was a fungal suspension containing 10^6 conidia ml^{-1} collected from 10-day-old PDA cultures of *Fon* races 0, 1, and 2. Seedlings of rootstocks and scions were grown in sterile sand. After removing the 15-day-old seedlings from the pots, the roots were washed with tap water and pruned to *ca* 1.0–1.5 cm. The roots were then dipped into inoculum solution for 3–4 min after which seedlings were transplanted into a tray of sterile sand. The number of healthy and dead plants in each genotype was recorded at 2-day intervals. Observations continued for 15 days (11). For grafted plants, rootstocks were grown in pots (8×8×8 cm) filled with a mixture of peat and perlite (2:1) and grafted at the first true-leaf stage. When grafted plants started to grow, they were inoculated with the pathogen by irrigating with 5 ml of conidial suspension. To facilitate inoculation, roots were first wounded with a spatula. Each treatment was replicated three times with five plants. Inoculated plants were kept at 24±1°C. Observations for healthy and dead plants continued for 20 days. In artificial inoculation in the laboratory, races 0, 1, and 2 defined in the Çukurova region by Yücel *et al.* (22) were used. In both inoculation tests, cv. Crimson Tide was used as control.

In the field, grafted and control plants were observed for signs of wilting. Root and stem tissue of plants affected by wilt symptoms was washed with running tap water and cut into pieces (3–4 mm) and sterilized with 1% NaOCl for 1–2 min. Plant samples were

TABLE 1. Names, properties and source of rootstocks

Rootstocks	Definition	Source
Cucurbita moschata (CMO)	Landrace	Mersin, Turkey
Cucurbita maxima (CMA)	Landrace	Adapazarı, Turkey
Lagenaria siceraria (LSC)	Landrace	Urfa, Turkey
Luffa cylindrica (LCY)	Landrace	Adana, Turkey
Benincasa hispida (BH)	Landrace	Ankara, Turkey
216	*Lagenaria* hybrid	Korea
Emphasis (EMP)	*Lagenaria* hybrid	Korea
Skopje (SKP)	*Lagenaria* hybrid	Korea
FR Gold (FRG)	*Lagenaria* hybrid	Korea
P360	*Cucurbita* hybrid	France
Strong Tosa (ST)	*Cucurbita* hybrid	Korea

rinsed with distilled water three times and excessive water on plant tissue was removed by sterile 3 MM Whatman filter paper. Disinfested plant pieces were cultured on PDA plates amended with 50 μg ml^{-1} streptomycin sulfate. The plates were incubated at 25±2°C for 7 to 10 days. Colonies exhibiting the taxonomic features of *Fon* were identified according to the procedures of Lecoq *et al.* (12) and Nelson *et al.* (16). Race definition was not done.

Crop conditions and plant sampling The grafted plants and the control plants were transplanted under low plastic tunnels; the tunnels were removed when the air temperature was suitable (22–25°C) for watermelon. Fertilizer was applied at the rate of 180 kg N ha^{-1}, 200 kg K_2O_5 ha^{-1} and 180 kg K_2O ha^{-1}. Micro-nutrients fertilization was not done. Total P was applied before planting. Nitrogen and potassium were divided into three equal portions and applied before planting, 20 days after planting and 40 days after planting. The experimental design was randomized complete blocks. Each treatment was replicated four times, with 15 plants in each replicate. Seedlings were transplanted in single rows spaced 2.0 m apart with 0.5 m between plants. Water was applied by drip irrigation. Fruits were harvested when ripe. Total yield (kg ha^{-1}) and marketable yield (%) were evaluated; five fruits from each replicate were randomly chosen to determine the fruit weight (g), fruit index (fruit length/fruit diameter), rind thickness (mm), flesh firmness (in Newtons, *N*), soluble solids, total and reduced sugar content of the juice (%). The selected fruits were sliced and rinds and seeds were removed. Juice was extracted from each fruit and soluble solids concentration (°Brix) was determined at 20°C using a hand refractometer. Firmness of the heart portion of the watermelon was determined with an Effegi Fruit Firmness Tester (Italy). This involved measuring the force required for an 8-mm probe to penetrate the cut surface to a depth of 5 mm at three locations in the mesocarp tissue. The total sugar content was determined using the sulfuric acid/phenol technique described by Dubois *et al.* (5). The reduced sugar content was determined by the dinitrophenol technique described by Rose (19). The study was performed over a 2-year period in 1999 and 2000.

Analysis of variance was performed using the COSTAT statistical program and means were compared using the Tukey test at the 1% significance level.

RESULTS AND DISCUSSION

Fusarium resistance The results of the *Fusarium* resistance tests and field observations are presented in Table 2. All rootstocks and grafted plants used in this study are resistant to three known races of *Fon* (11,12). Watermelon cv. Crimson Tide is susceptible to race 2. In field observations, wilting caused by *Fon* was not observed in any of the grafted plants but was observed in 5–10% of the control plants. *Fon* spores and micelles were found in the vascular bundles of wilted plants.

TABLE 2. Resistance of rootstocks, grafted and ungrafted Crimson Tide (CT) watermelon cultivar (scion) to three races of *Fusarium oxyporum* f.sp. *niveum*

Rootstocks and grafted plant		Mode of inoculation[z]	Fusarium races			Field obser-vation
			0	1	2	
CMO	Rootstock	Root dip	R[y]	R	R	
CT/CMO	Grafted	Irrigation	R	R	R	–
CMA	Rootstock	Root dip	R	R	R	
CT/CMA	Grafted	Irrigation	R	R	R	–
LCY	Rootstock	Root dip	R	R	R	
CT/LCY	Grafted	Irrigation	R	R	R	–
BH	Rootstock	Root dip	R	R	R	
CT/BH	Grafted	Irrigation	R	R	R	–
P360	Rootstock	Root dip	R	R	R	
CT/P360	Grafted	Irrigation	R	R	R	–
ST	Rootstock	Root dip	R	R	R	
CT/ST	Grafted	Irrigation	R	R	R	–
216	Rootstock	Root dip	R	R	R	
CT/216	Grafted	Irrigation	R	R	R	-
FRG	Rootstock	Root dip	R	R	R	
CT/FRG	Grafted	Irrigation	R	R	R	-
EMP	Rootstock	Root dip	R	R	R	
CT/EMP	Grafted	Irrigation	R	R	R	-
SKP	Rootstock	Root dip	R	R	R	
CT/SKP	Grafted	Irrigation	R	R	R	-
LSC	Rootstock	Root dip	R	R	R	
CT/LSC	Grafted	Irrigation	R	R	R	-
CT (control)	Scion	Root dip /Irrigation	R	R	S[x]	+

[z] See Materials and Methods.
[y] R= resistant.
[x] S= Susceptible.
– Wilting caused by *Fusarium* was not observed in the field.
+ Wilting caused by *Fusarium* was observed in the field.

The vigorous roots of the rootstocks exhibited superior tolerance to serious soilborne diseases, such as those caused by *Fusarium* (9) and *Verticillium* (7). Disease resistance in grafted plants may be attributed entirely to the tolerance of the roots of the stock plants to such diseases (12). *Fon* is host-specific for watermelon and therefore none of the rootstocks was infected. Either the pathogen could lack a specific pathogenicity function required to cause disease in the host, or the host could lack a specific susceptibility factor (20). Biles *et al.* (2) reported that a substance associated with *Fusarium* resistance was synthesized in the root and translocated to other parts of the plant through the xylem.

Yield and fruit quality *Benincasa hispida* and *Luffa cylindrica* LCY did not emerge in early spring due to low temperatures. Therefore, grafted seedling material could not be produced for yield and quality experiments in these two rootstocks (Table 3). Yield appeared to be significantly affected by rootstocks. Plants grafted onto *Lagenaria*-type rootstocks (216, FRG, EMP, SKP, LSC) produced higher yields than the control and *Cucurbita*-type rootstocks (CMO, CMA, ST, P360). The highest yield was obtained from CT/SKP and

TABLE 3. Yield and quality characteristics of watermelon grafted onto different rootstocks (CT, Crimson Tide = control

Rootstocks	Total Yield (kg ha^{-2})	Marketable Yield (%)	Fruit Weight (g)	Fruit Index	Rind Thickness (mm)	Flesh Firmness (Nt)	TSS (%)
CT/CMO	22,910 e[z]	78.5 ab	2034 c	1.23	10.73 b	12.63 a	7.49 b
CT/CMA	19,530 e	50.5 c	3142 c	1.05	10.55 b	11.62 a	8.05 b
CT/LCY[y]							
CT/BH[y]							
CT/P360	18,910 e	59.5 bc	3310 c	1.10	16.72 a	8.13 b	9.01 ab
CT/ST	18,760 e	71.5 abc	7487 ab	1.15	16.33 ab	7.44 b	9.60 a
CT/216	83,790 cd	95.8 a	7260 ab	1.11	15.89 ab	6.36 b	9.76 a
CT/FRG	89,600 c	94.8 a	7577 ab	1.16	13.57 ab	7.05 b	9.29 a
CT/EMP	109,520 b	95.3 a	7233 ab	1.19	14.02 ab	7.64 b	9.50 a
CT/SKP	146,430 a	97.0 a	8809 a	1.20	16.23 ab	8.03 b	9.30 a
CT/LSC	136,940 a	96.8 a	7346 ab	1.16	14.05 ab	7.34 b	9.07 ab
CT	68,970 d	83.0 ab	5854 bc	1.21	11.78 ab	6.85 b	9.46 a
MSD (1%)	15,470	25.88	2048	0.28	5.94	2.84	1.78

[z]Within columns, figures followed by a common letter do not differ significantly at $P \leqslant 0.01$.
[y]The emergence rate was not sufficient for grafting in early spring.

TABLE 4. Reduced and total sugars contents of watermelon fruit juice (CT, Crimson Tide = control)

Combinations	Reduced Sugar (%)	Total Sugar (%)
CT/CMO	4.74 c[z]	7.22 bc
CT/CMA	4.46 c	6.92 c
CT/P360	4.92 bc	8.62 a
CT/ST	5.85 abc	8.60 a
CT/216	5.17 abc	8.62 a
CT/FRG	5.87 abc	8.90 a
CT/EMP	5.41 abc	8.30 ab
CT/SKP	6.33 ab	8.37 ab
CT/LSC	6.51 a	8.97 a
CT	5.61 abc	8.90 a
MSD (1 %)	1.57	1.36

[z]Within columns, figures followed by a common letter do not differ significantly at $P \leqslant 0.01$.

CT/LSC combinations, with 146,430 kg ha^{-1} and 136,940 kg ha^{-1}, respectively. While control plants yielded 68,970 kg ha^{-1}, the plants grafted onto *Cucurbita*-type rootstocks produced about 20,000 kg ha^{-1} yield (Table 3). Marketable yield was also influenced by rootstock. The plants grafted onto *Lagenaria*-type rootstocks produced higher marketable yields than control and *Cucurbita*-type rootstocks. Incompatibility observed in the latter rootstocks caused the low total and marketable yields in the plants grafted onto *Cucurbita*-type rootstocks. Since fruits on the Crimson Tide/*Cucurbita*-type rootstock could not attain their original fruit size, and yield was low (Table 3).

Many authors have stated that a rootstock promoted higher yields in grafted plants (3,17,20). These increases can be explained by an interaction of some or all of the following phenomena: increased water and plant nutrient absorption (10), augmented endogenous hormone production (24), and enhanced scion vigor (8,14), resistance to soil pathogens (6,13), tolerance to low soil temperature (4,21) and to salinity (23). In our study, fruit yield was significantly affected and increased by rootstocks except where there was an incompatibility problem. In those combinations, small fruit size and low fruit quality occurred because of incompatibility. Incompatible plants died during the fruit production stage of growth and therefore fruit ripened before it could attain its original size and quality. Joint effects of the processes mentioned above can explain yield increases.

Fruit characteristics of watermelon grafted onto different rootstocks are presented in Table 3. Plants grafted onto *Lagenaria*-type rootstocks produced larger fruit than control plants and plants grafted on *Cucurbita*-type rootstocks. The biggest fruit was obtained from the CT/SKP combination with a weight of 8809 g, whereas fruit from the control plants weighed 5854 g. The plants grafted onto *Cucurbita*-type rootstocks weighed ~3000 g. Fruit index (shape) was not significantly affected by the rootstock but rind thickness was affected, with *Lagenaria*-type rootstocks resulting in thicker fruit rinds, although the difference was not economically important. Bigger fruit had thicker rinds. Flesh firmness was significantly affected by rootstocks, the greatest firmness being found in the CT/CMO and CT/CMA combinations: 12.63 N and 11.62 N, respectively. The control and other combinations had lower values (6.36–8.13 N) and were statistically similar. The soluble solids contents were significantly affected by the rootstock. All rootstocks gave similar results in regard to soluble solids except CMO (7.49%) and CMA (8.05%), which evinced severe incompatibility with watermelon. About 50% of the plants wilted and then died at the fruit enlarging period due to incompatibility. Swelling was observed at the graft site in incompatible combinations.

Reduced and total sugars contents of watermelon juice are presented in Table 4. The reduced sugar content varied from 4.46% to 6.51% and was significantly affected by the rootstock. The highest reduced sugar content was found in the CT/LSC combination (6.51%) and the lowest in CT/CMO and CT/CMA (4.74% and 4.46%, respectively).

Total sugar content of watermelon juice varied with the rootstock. The highest total sugar content was found in the CT/LSC combination (8.97%) and the lowest in CT/CMA (6.92%). Control and grafted plants had similar total sugar contents except for the CT/CMA and CT/CMO combinations. Contrary to another report (13), detrimental effects of rootstock on fruit quality were not found in grafted plants with the exception of CT/CMO and CT/CMA combinations.

In conclusion, our observations with watermelon show that grafting seems to be an effective tool for disease resistance as well as having a positive effect on yield and quality. *Lagenaria*-type rootstocks were resistant to three known races of *F. oxysporum* f.sp. *niveum* and were also capable of significantly improving productivity without exhibiting any detrimental effects on fruit quality of the cultivars used as scions. Skopje (SKP) and *Lagenaria siceraria* (LSC) can be considered as the most suitable rootstocks for watermelon. The landrace LSC is recommended as a source of breeding material for rootstocks of watermelon.

ACKNOWLEDGMENTS

This research was supported by Project No. TOGTAG TARP-2410 from The Scientific and Technical Research Council of Turkey (TUBITAK), Ankara, Turkey.

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