Percent of Survival, Vegetative Growth and Hormonal Balance as Affected by Chip Budding of Flame Seedless cv. on Three Nematode Resistant Rootstocks

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ABSTRACT
The present study was carried out during two successive seasons of 2012 and 2013 in a private vineyard located at Sadat city, Menofia Governorate, Egypt. The main target is the detection of the proper rootstock which achieve the highest compatibility (Congeniality) degree for the Flame seedless cv., with good survival % and healthy growth for the obtained grafts especially vegetative growth of scion and root system growth of the rootstock. The survival percentage varied significantly according to rootstock type. Freedom rootstock recorded the highest survival percentage (80.00 and 83.33 %) and F.C.C (2.41 and 2.53) in 2012 and 2013 seasons, respectively. While, Harmony rootstock recorded the lowest survival percentage (60.00 and 66.67 %) and F.C.C (1.51 and 1.82) in both seasons, respectively. Meanwhile, difference in F.C.C. value between Freedom and Salt creek was insignificant in both seasons. Freedom rootstock recorded the maximum values of scion shoot length (93.55 and 95.86 cm) and leaf area (95.24 and 105.49 cm2) in 2012 and 2013 seasons, respectively. While, Harmony rootstock achieved the minimum values of shoot length (67.43 and 71.67 cm) and leaf area (73.37 and 79.60 cm2) in the two studied seasons, respectively. However, differences in both shoot length and between Freedom and Slat creek rootstocks are insignificant in both seasons. Freedom rootstock recorded the maximum figures of the aerial portion fresh weight (7.65 and 8.89 g), fresh weight of root system (2.94 and 4.23 g), dry weight of the aerial portion (2.07 and 2.52 g) and dry weight of root system (1.7 and 2.4 g) in the two seasons, respectively. While, Harmony rootstock showed the minimum values of fresh weight of aerial portion (5.65 and 6.33 g), fresh weight of root system (2.30 and 2.91 g), dry weight of the aerial portion (1.65 and 1.81 g) and dry weight of root system (1.34 and 1.63 g) in the two studied seasons, respectively. Meanwhile, differences in fresh wt. and dry wt. of aerial portion between Freedom and Salt creek were insignificant in both seasons. The obtained results revealed that canes of Freedom rootstock exhibited the largest total indoles (0.29 and 0.30 mg/g F.W.), free indoles (0.25 and 0.26 mg/g F.W.). But they showed the lowest total phenols (0.17 and 0.15 mg/g F.W.) and free phenols (0.15 and 0.13 mg/g F.W.) in 2012 and 2013 seasons, respectively. On the contrary, canes of Harmony rootstock exhibited the minimum values of total indoles (0.24 and 0.23 mg/g F.W.) and free indoles (0.19 and 0.20 mg/g F.W.) and the highest figures of total phenols (0.24 and 0.23 mg/g F.W.) and free phenols (0.20 and 0.18 mg/g F.W.) in both seasons, respectively. However, no significant differences were detected in total indoles and total phenols between Freedom and Slat creek rootstock in both seasons.

Keywords: Chip budding, Flame seedless, Nematode

1. Introduction
Grape (Vitis vinifera L.) is one of the most important fruit crops on a worldwide because it is the fourth crop all over the world after citrus, apple and bananas. Turkey is the world's largest grape producer followed by The United States of America and then China. Grapevine (Vitis vinifera L.) which included all edible cultivars is nematode sensitive. Nematodes cause serious troubles on grapevines...
such as root gall formation, general weakness, reduced yield and transmission of viral agents. Therefore, grafting on nematode resistant rootstocks could be a powerful solution to avoid infection with nematodes. Recently in the last decades, grafting became gradually the common way to propagate the promising cultivars of grape. In this concern, some nematode resistant grape rootstocks i.e. Freedom, Harmony, Salt creek, SO4, Richter...etc had been enrolled in Egypt.

The Harmony rootstock was developed by Harmon and Weinberger at the USDA-Fresno and it is a cross between seedling of Dog ridge and a seedling of 1613. Freedom rootstock is a similar cross but the seedling selections were different and it was selected in 1956. Salt creek rootstock correctly named Ramsey rootstock, because there is another variety (American Hybrid) called Salt creek rootstock, which is a selection of V. champinii. Much like Dog ridge except easier to manage. Flame seedless grape is one of the most common grape cvs. and it is an early season cultivar (Mahmoud, 2010). Mikhail (1976) found that the survival percentage of Thompson Seedless and Italia cvs. that had grafted on various rootstocks differed according to the rootstocks used, grafting date and grafting method. Moti Singh and Chaudhry (1948) reposed that the survival percentage was the greatest (100%) when grafting Pearl of Casba cv. On Kandhari rootstock followed by Bharat Early cv. grafted on Kandhar rootstock (93.3%) and was (83.3%) on Gulongri rootstock. A study was carried by Bhujbal (1993) stated that high survival percentage was recorded when grafting Thompson seedless on P-1103.

The survival percentage of 8 cultivars grafted on 7 different rootstocks had been investigated. The highest survival percentage was recorded for those grafted on Kober 5BB rootstock (84.9%), and the lowest survival percentage was in vines grafted (71.2 %) on Vitis solonis × V. riparia 1616 (Hubackova, 1994). The scion vegetative growth is affected by using different rootstocks. Sauer (1972) reported that Sultana grapes vines grafted on two V. champinii stocks were more vigorous than other vines, where the vegetative growth was greater on Dog Ridge than on Salt Creek. Raski et al., (1973) noted that Dog Ridge and Salt creek rootstocks grafted with Granech scions were equally vigorous and productive. Crescimanno et al., (1981) showed that the growth vigor of own rooted cuttings compared with grafting on different rootstocks. Grapindaivili and Tsertsevaoze (1981) found that the grapevine cv. Krakhuna had the largest leaf area when grafted on Chasselas x Berlandieri rootstock. Mortensen and Stover (1982) reported that the scion vigour was greater on Tampa than Dog Ridge, Lake Emerald or Blue Lake.

Pongrancy (1983) studied the effect of grafting on leaf area and observed that there was a great variation in leaf area of scions due to different grape rootstocks. Hedberg et al., (1986) demonstrated that the vegetative growth was increased of Shiraz cv. grafted on Ramsey and Dog Ridge rootstocks. Scienza et al., (1986) stated that some rootstocks are producing high amount of dry matter. Ruhl (1991) reported that dry matter production was not affected by some rootstocks. El-Hawary (1987) found that the largest shoots were obtained when Romi Red cultivar has grafted on ARG rootstock, while the shortest shoots was obtained when grafted on Couderc 1202 rootstock. On the other hand, the largest leaf area of Romi Red cultivar was recorded when grafted on Berlandieri x Reparia rootstock and the smallest leaf area obtained when grafted on ARG rootstock. Somkuwar and Adsule (2009) when grafted Thompson Seedless on four grape rootstocks (Dog Ridge, Salt Creek, 1613-C and St. George. They found the dry matter was high in Thompson Seedless grafted on Dog Ridge (69.48%), while St. George was last in the order (43.79%). Canes of Salt Creek grafted vines produced highest dry matter (73.57%). However less dry matter was recorded in roots of 1613-C grafted vines.

The main target was detection of the proper rootstock which achieve the highest compatibility (Congeniality) degree for the Flame seedless cv, with good survival % and healthy growth for the obtained grafts especially vegetative growth of scion and root system growth of the rootstock.

2. Material and Methods

The present study was carried out during two successive seasons of 2012 and 2013 in a private vineyard located at Sadat city, Menofia Governorate, Egypt to investigate the influence of three nematode resistant grapevine rootstocks; namely, Harmony, Freedom and Salt creek grafted with Flame seedless scions on August 15th in both seasons using the Chip technique mainly to find out the proper rootstock grafting.
Callusing was done after budding in both seasons of study by using the callusing box which kept at 85-90 % R.H and 25-28° C and when the grafts achieved 75% success in good callus formation covering the grafting union the practice was terminated.

Each graft was planted in black polyethylene bag 19.5×11.5×9.5 cm. containing peat moss and sand mixture (2:1v/v). The polyethylene bags were placed under plastic tunnels (60 micron) till the end of April after which the cover was removed and the usual irrigation and management were practiced. Each treatment replicated 3 times and each replicate was comprised of 10 cuttings.

2.1. Considered parameters
a) Survival percentage
   The survival percentage (percentage of sprouted grafts with rooted Rootstocks) was estimated after one month of transplanting.

b) Growth parameters of the scion and rootstocks.
   1. Shoot length (cm): after six months of transplanting.
   2. Leaf area (cm2).
      Leaf area of the fifth apical full expanded leaf after six months of transplanting was measured using the following equation: Leaf area = 3.14 × square diameter /4 according to Sorial et al. (1985).
   3. Fresh weight of aerial portion (leaves and shoots) (g) on mid. Oct.
   4. Fresh weight of root system (g) of rootstocks on mid. Oct.
   5. Dry weight of aerial portion (leaves and shoots) (g) on mid. Oct.
   6. Dry weight of root system (g) of rootstocks on mid. Oct.

c) Total indoles and free indoles were determined for samples were taken from grafting union zone before grafting date. About 0.5 g of fresh samples were mixed with 25 ml of 50% methanol/water and heated at 40 °C in a plastic screw-capped tube with intermittent shaking for 1 h to determine the free indoles. Another 0.5 g of dry weight samples were heated with 25 ml of 1.2 M HCl in 50% aqueous methanol and Para-dimethyl amino benzialhyed for 1 h at 40 °C to measure the total phenols. The both total indoles and free indoles were determined calorimetrically at 530 nm wave length and the concentrations calculated as mg. indole acetic acid per g. dry weight according to Selim et al. (1978).

d) Total phenols and free phenols were determined for samples were taken from grafting union zone before grafting date. About 0.5 g of fresh samples were mixed with 25 ml of 50% methanol/water and heated at 90 °C in a plastic screw-capped tube with intermittent shaking for 2 h to determine the free phenols. Another 0.5 g of dry weight samples were heated with 25 ml of 1.2 M HCl in 50% aqueous methanol for 2 h at 90 °C to measure the total phenols. Total and free phenols were determined by the Folin- Ciocalteu assay at 370 nm wave length. The concentration was calculated from a standard curve of phrogallol as mg 100g dry weight (Singleton et al., 1999).

Statistical analysis
   In the experiment, one grafting date was used only for the three rootstocks in a factorial design rootstocks. Each one was represented by 3 replicates where each one comprised 10 grafts. The statistical analysis of the present data was carried out using Analysis of Variance (ANOVA) according to Snedecor and Chocran (1990). The averages were compared using Duncan Multiple range test (Duncan, 1955) at 5 % level.

3. Results and Discussion
   Data in Table (1) show that the survival percentage varied significantly according to rootstock type. Freedom rootstock recorded the highest survival percentage (80.00 and 83.33 %) and F.C.C (2.41 and 2.53) in 2012 and 2013 seasons, respectively. While, Harmony rootstock recorded the lowest survival percentage (60.00 and 66.67 %) and F.C.C (1.51 and 1.82) in both seasons, respectively. Meanwhile, difference in F.C.C. value between Freedom and Salt creek was insignificant in both seasons.
Table 1: Survival percentage and field compatibility constant (FCC) of Flame seedless scion grafted on the three studied rootstocks with Chip Budding method in mid-August of 2012 and 2013 seasons.

| Grafting date | Rootstocks | 2012 | 2013 |
|---------------|------------|------|------|
| Survival (%)  | Survival (%)| Field compatibility constant | Field compatibility constant |
| Freedom       | 80.00 A    | 2.41 A | 83.33 A | 2.53 A |
| Harmony       | 60.00 C    | 1.51 B | 66.67 C | 1.82 B |
| Salt creek    | 66.67 B    | 2.10 A | 73.33 B | 2.24 A |

Means having the same letter(s) in each column or row are in significantly different at 5% level.

3.1. Vegetative growth parameters of the scion.

Data in Table (2) indicated that shoot length and leaf area of scion grafted on the three studied rootstocks with Chip budding method in Mid-August of 2012 and 2013 seasons significantly varied according to rootstock type. Freedom rootstock recorded the maximum values of scion shoot length (93.55 and 95.86 cm) and leaf area (95.24 and 105.49 cm²) in 2012 and 2013 seasons, respectively. While, Harmony rootstock achieved the minimum values of shoot length (67.43 and 71.67 cm) and leaf area (73.37 and 79.60 cm²) in the two studied seasons, respectively. However, differences in both shoot length and between Freedom and Salt creek rootstocks are insignificant in both seasons.

Table 2: Shoot length (cm.) and leaf area (cm²) of Flame seedless scion grafted on the three studied rootstocks with Chip Budding method in Mid-August of 2012 and 2013 seasons.

| Grafting date | Rootstocks | 2012 | 2013 |
|---------------|------------|------|------|
| Shoot length (cm.) | Leaf area (cm²) | Shoot length (cm.) | Leaf area (cm²) |
| Freedom       | 93.55 A    | 95.24 A | 95.86 A | 105.49 A |
| Harmony       | 67.43 B    | 73.37 C | 71.67 B | 79.60 C |
| Salt creek    | 85.48 AB   | 83.08 B | 88.45 AB | 86.21 B |

Means having the same letter(s) in each column or row are in significantly different at 5% level.

Data in Table (3) illustrated that Freedom rootstock recorded the maximum figures of the aerial portion fresh weight (7.65 and 8.89 g), fresh weight of root system (2.94 and 4.23 g), dry weight of the aerial portion (2.07 and 2.52 g) and dry weight of root system (1.7 and 2.4 g) in the two seasons, respectively. While, Harmony rootstock showed the minimum values of fresh weight of aerial portion (5.65 and 6.33 g), fresh weight of root system (2.30 and 2.91 g), dry weight of the aerial portion (1.65 and 1.81 g) and dry weight of root system (1.34 and 1.63 g) in the two studied seasons, respectively. Meanwhile, differences in fresh wt. and dry wt. of aerial portion between Freedom and Salt creek were insignificant in both seasons.

Table 3: Fresh weight of the aerial portion and root system (g) and dry weight of the aerial portion and root System (g) of Flame seedless scion grafted on the three studied rootstocks with Chip Budding method in mid-August of 2012 and 2013 seasons.

| Grafting date | Rootstocks | 2012 | 2013 |
|---------------|------------|------|------|
| F.W. of the aerial portion (g) | F.W. of root system (g) | D.W. of the aerial portion (g) | D.W. of the root system (g) |
| Freedom       | 7.65 A    | 2.94 A | 2.07 A | 1.65 A | 8.89 A | 4.23 A | 2.52 A | 2.41 A |
| Harmony       | 5.65 B    | 2.30 B | 1.65 B | 1.34 C | 6.33 B | 2091 C | 1.81 B | 1.63 C |
| Salt creek    | 7.02 A    | 2.86 A | 1.92 AB | 1.59 B | 7.16 AB | 3.57 B | 2.20 AB | 2.09 B |

Means having the same letter(s) in each column or row are in significantly different at 5% level.
Table (4) revealed that canes of Freedom rootstock exhibited the largest total indoles (0.29 and 0.30 mg/g F.W.), free indoles (0.25 and 0.26 mg/g F.W.). But they showed the lowest total phenols (0.17 and 0.15 mg/g F.W.) and free phenols (0.15 and 0.13 mg/g F.W.) in 2012 and 2013 seasons, respectively. On the contrary, canes of Harmony rootstock exhibited the minimum values of total indoles (0.24 and 0.23 mg/g F.W.) and free indoles (0.19 and 0.20 mg/g F.W.) and the highest figures of total phenols (0.24 and 0.23 mg/g F.W.) and free phenols (0.20 and 0.18 mg/g F.W.) in both seasons, respectively. However, no significant differences were detected in total indoles and total phenols between Freedom and Slat creek rootstock in both seasons.

Table 4: Total indoles, free indoles, total phenols and free phenols (mg / g F.W.) in canes of various studied rootstocks in mid August 2012 and 2013 seasons

| Grafting date | Rootstocks | Total Indoles (mg/g F.W.) | Free Indoles (mg/g F.W.) | Total phenols (Mg/g F.W.) | Free phenols (Mg/g F.W.) |
|---------------|------------|--------------------------|--------------------------|--------------------------|--------------------------|
| 2012          | Freedom    | 0.29A                    | 0.25A                    | 0.17B                    | 0.15B                    |
|               | Harmony    | 0.24A                    | 0.19B                    | 0.24A                    | 0.20A                    |
|               | Salt creek | 0.27A                    | 0.22AB                   | 0.18B                    | 0.17AB                   |
| 2013          | Freedom    | 0.30A                    | 0.26A                    | 0.15B                    | 0.13C                    |
|               | Harmony    | 0.23B                    | 0.20B                    | 0.23A                    | 0.18A                    |
|               | Salt creek | 0.28AB                   | 0.23B                    | 0.17B                    | 0.14AB                   |

Means having the same letter(s) in each Column or row are in significantly different at 5% level.

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