



The Effect of Grafting on Squash Plants Grown Under Low Plastic Tunnel in Winter Season



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THIS study was carried out during the two successive seasons of 2015-2016 and 2016-2017 at Kaha Research Station, Horticulture Research Institute (HRI), Agricultural Research Center (ARC), Ministry of Agriculture, Egypt under low plastic tunnel to study the effect of grafting to resist low temperature in winter season the effect of grafting on vegetative growth, yield, and fruit quality of two squash hybrids (*Cucurbita pepo* L.) 'Aziad and Raad'. Squash transplants were grafted on three rootstocks namely *Lagenaria siceraria* Standl (Lg), *Cucurbita ficifolia* (Cf), *Cucurbita moschata* (Cm) and self-grafted squash transplants used as a control. All transplants were grown under low plastic-tunnel conditions. The results showed that vegetative growth characters i.e., plant height, number of leaf per plant and leaf area were significantly increases when 'Aziad' hybrid was grafted on *Cucurbita moschata* (Cm) in both seasons. The same results were obtained with the characters of fruit quality i.e. number of marketable fruits/plant, marketable fruit weight/plant, V.C., carbohydrates, free proline and total chlorophyll. While, there were no significant differences realized when Aziad or Raad' grafted on the others rootstocks (Lg and Cf) compared with self-grafted plants (control) on leaves dry weight, average of fruit weight, Ca%, dry weight of 100g fresh fruit, TSS and total carotenoids.

Keywords: Squash, Grafting, Rootstocks, Low plastic tunnel, Vegetative growth, Fruit yield and quality.

Introduction

Squash (*Cucurbita pepo* L.) is one of the most important and popular vegetable crop in the world. Squash's family is one of the largest families in the plant kingdom consisting of largest number of plant species supplying humans with edible products and useful fibers (Smith, 1997). Vegetable constitutes are an important component in human's diet, especially in developing countries, it was supplying essential minerals and vitamins that may not be obtained solely from staples. Squash plant produces a lot of biomass and its nutrient requirements are generally considered to be high particularly nitrogen and phosphorus (Obalum et al., 2012). Most known plants multiply from seeds (sexual) whereas certain plants are preferentially multiplied from their

parts such as stem, roots, or leaves. Multiplication of plants using parts other than seeds was known as vegetative (asexual) propagation and the resultant plants are referred to as clones. For various reasons, some plants are multiplied by combining vegetative plant parts (stem or vegetative buds) from two separate plants into one. Grafting and budding are techniques used to combine one plant part with another to encourage growth as a unified plant (Kumar, 2011). Grafting technique is currently practiced worldwide on many high-value cucurbitaceae (watermelon, melon, and cucumber) and solanaceae (tomato, eggplant, and pepper) crops for both open-field and protected cultivations (Davis et al., 2008, Lee & Oda, 2003 and Lee et al., 2010). Grafting is applied in the agricultural practice for protection

of plants against plant disease and environmental adverse in cucurbitaceae family. Also grafting Synthesized suitable between roots and shoots to generate chimeras that are more vigorous, more pathogen resistant and more abiotic stress resistant (Melnyk 2017). Grafting of cucumber on four different rootstocks namely Bottle Gourd (*Lagenaria siceraria* Standl.), Supper Shintosa (*Cucurbita maxima* Duchesne × *Cucurbita moschata* Duchesne), Squash 3 (*Cucurbita pepo*) and Ferro (*C. maxima* × *C. moschata*) resulted in an increase in cucumber plant height when grafted on Ferro rootstock in both summer and winter seasons, compared with (control) and other rootstocks used (El-Sayed et al., 2014). Rootstocks considered as powerful of supply water and nutrient uptake by the soil leads to distribute growth regulators to the grafted parts (Helaly, 2017). The effect of grafting on cucumber, watermelon and melon plants grafted onto *Cucurbita* spp. rootstocks. Three rootstocks were used: (*Cucurbita pepo* L.), (*Cucurbita moschata* L.) and (*Cucurbita ficifolia* Bouche). Survival rate of grafted plants was significantly affected by rootstock. The grafting was highly effective in watermelon and cucumber, (*Cucurbita pepo* L.) and (*Cucurbita moschata* L.) were used as rootstocks. Fig-leaf rootstock was not found highly effective even for cucumber with grafting. Corresponding rates for melon were found lower, especially with the fig-leaf rootstock (Salehi et al., 2008). Grafting is an important technique tool to improve vegetable growth, yield production and fruit quality (Marsic & Jakse 2010, Schwartz et al., 2010, and Farhadi et al., 2016). In other study grafted cucumber cv Gianco RZ fl hybrid on different rootstocks. Various 6 genotypes of rootstocks from cucurbitaceae were used for grafting: Gordal, Luffa, Pumpkin, Bottle gourd, Star and Hersh in addition to Control (self-grafted plant). The highest leaves number was revealed from Star rootstock compared to the control, while the biggest leaf area was obtained from Botle gourd rootstock compared to Luffa rootstock which gave smallest leaf area. On the other side the highest fruit total yield was obtained from Star rootstock. The physical and chemical traits of grafted cucumber fruits effected significantly on the changes of scion product after grafting. Overall, its recommended that the use of Hersh and Star rootstocks could be provide a useful tool to improve vegetative growth (Alaeldin et al., 2019).

This work aimed to study the effect of grafting squash transplants on different rootstocks on growth, yield and some fruit quality characters of squash plants grown under low plastic tunnel conditions to resist low temperature in winter season.

Materials and Methods

This experiment was carried out under low plastic tunnel at Kaha Research Station, Horticulture Research Institute (HRI), Agricultural Research Center (ARC), Ministry of Agriculture, Giza Governorate, Egypt during two successive growing seasons of 2015-2016 and 2016-2017. Squash hybrids Aziad and Raad 'transplants' were grafted onto three rootstocks namely *Lagenaria siceraria* (Lg), *Cucurbita ficifolia* (Cf), *Cucurbita moschata* (Cm) as well as self-grafted squash hybrid transplants were used as control. Seeds of rootstocks as well as seeds of squash hybrids were sown in Styrofoam seedling trays (84 cells) filled with a mixture of growing media peat moss: vermiculite (1: 1V/V), supplemented with 50 g of a fungicide for each 50 kg of the mixture media peat moss : vermiculite (1:1 V/V). Seeds of used rootstocks Lg, Cf and Cm were sown on 5th of November, while scions seeds squash hybrid 'Aziad' and 'Raad' were sown on 12th of November with one week interval on both seasons of study. Then the trays were kept in a greenhouse and cared by regular practices for seedlings production under greenhouse conditions, after 18 days from seed sowing the stems of scions and rootstocks were cut at right angles with leaving 2 to 3 true leaves depending on prevailing temperature. The tapered stems of the scions were placed into the cleft of the cut-end of the rootstocks, followed by clipping. It was important during grafting to increase the chance for vascular bundles of the scions and rootstocks to come into contact. This was achieved by increasing the area of spliced cut surfaces and by appropriate pressure to the spliced cut surfaces together should not be dried (Oda et al., 1995). Then the grafted seedlings were placed under a plastic tunnel to provide an optimum temperature between 25-30°C by using (digital thermal/hygrometer Art. No.30.5000/30.5002, TFA, Germany) and humidity (98%). After seven days from grafting stage by watching the new growth on the scions. The plastic tunnel was gradually opened for adaptation and preparing the grafted seedlings for transplanting in the low plastic tunnel, where the basic experiment was carried

out. Grafted squash transplants were transplanted on 15th December in the first and second seasons. The split plot experimental design was used with three replicates where the two hybrids arranged in the main plots, and the rootstock treatments were randomly arranged in the sub plots.

The physical and chemical characteristics of the soil of the experimental are set out in Table 1

Measurements

Vegetative growth

After 90 days from transplanting date, a random sample of three plants from each replicate was taken and some vegetative growth parameters were recorded as follows: Plant height (cm), number of leaf per plant, leaf area (cm²) using leaf area meter (Portable Leaf Area Meter, MLA, China), leaf total chlorophyll (SPAD reading, TYS-A, ZHE JIANG TOP INSTRUMENT CO., LTD, China) was determined in the fully expanded leaf, the fourth plant leaf from the top of plant and leaves dry weights (g).

Physical characters of fruits

Data of all over the harvesting season per each replicate were recorded for squash fruits harvested at maturity stage and examined for the following characters: number of marketable fruits/plant, fruit weight/plant, average of fruit weight (g). Also a random sample of 10 fruits from each replicate was harvested to estimate the dry weight of 100 g fruit fresh weight (g).

Chemical characters of leaves and fruits

A random sample of 10 fruits from each replicate were harvested at maturity stage to determine the

following characters: total soluble solids (TSS), Calcium (Ca %) Ascorbic acid content (V.C.) and total carbohydrates (mg/g dry matter) as mentioned in A.O.A.C. (1990). Total carotenoids were determined by Using a volumetric pipette, transfer 2 ml (VF) of Solution D (see above) to an amber 100-ml (VG) volumetric flask. Add 10 ml of ethanol bring to volume with petroleum ether and mix well. This is sample Solution G. Using a suitable UV/VIS spectrophotometer and 1-cm sample cells with covers, scan the spectrum of Solution G from 550 to 300 nm, using petroleum ether as the reference blank and measure the absorbance at the absorbance maximum (approximately 472 nm). The absorbance should be between 0.2 and 0.8. Calculate the percentage of total carotenoids (as lycopene) in the sample using the following equation:

$$\text{TOTAL CAROTENOIDS \%} = (A \times D / W_s \times 3450) \times 100$$

A is the absorbance of Solution G at 472 nm, corrected for the blank, 3450 is the specific absorbance (A 1%1 cm) of all-trans-lycopene in petroleum ether, WS is the weight of the sample (g), and D is the dilution factor (VGxVC/VFxB). Free proline content was determined according to the method of (Bates et al., 1973).

Statistical analysis

Statistical analysis was performed using the generalized linear model (GLM) procedure of Gen STAT software. Least significant difference (LSD) at 0.05 level of probability was used to compare the significant differences among treatment means as described by Senedecor and Cochran (1989).

TABLE 1. Soil Characteristics of the Experimental Area.

| Characteristics | Season of 2015 | Season of 2016 |
|-----------------------|----------------|----------------|
| pH | 7.62 | 7.45 |
| ECe (ds/m) | 6.79 | 7.08 |
| Sand (%) | 25.7 | 28.9 |
| Salt (%) | 27 | 28.3 |
| Clay (%) | 46 | 44.1 |
| Organic Matter (%) | 1.02 | 0.96 |
| CaCO ₃ (%) | 3.68 | 3.85 |
| Texture class | CL | CL |
| Available N (ppm) | 198 | 242 |
| Available P (ppm) | 1.92 | 4.25 |
| Available K (ppm) | 161 | 265 |

Results and Discussion

The effect of squash hybrids

Data in Table (2) showed that grafting Aziad hybrid onto different rootstocks resulted in significant increases in plant height. As for number of leaves per plant there are no significance differences was realized between Aziad and Raad when used them as scions onto

different rootstocks in both seasons. There were no significance differences detected between Aziad and Raad concerning leaves dry weight in the first season only. Generally, Aziad hybrid recorded a higher leaves dry matter compared with Raad hybrid in both seasons of the study. While, the results for the leaf area showed no significance differences between the both investigated hybrids in the two seasons.

TABLE 2. The effect of different rootstocks on plant height, number of leaves per plant, leaves dry weight and leaf area of squash plants grown under low plastic tunnel in winter season during seasons of 2015/2016 and 2016/2017.

| Measurements | Plant height (cm) | | Number of leaves/plant | | Leaves dry weight (g) | | Leaf area (cm ²) | | |
|--|------------------------|------------------------|------------------------|------------------------|------------------------|------------------------|------------------------------|------------------------|--------|
| | 1 st season | 2 nd season | 1 st season | 2 nd season | 1 st season | 2 nd season | 1 st season | 2 nd season | |
| a) The effect of squash hybrids | | | | | | | | | |
| Aziad | 174.54 | 157.62 | 30.58 | 30.02 | 48.54 | 41.31 | 302.13 | 295.98 | |
| Raad | 142.51 | 112.19 | 24.88 | 26.38 | 42.33 | 34.39 | 288.86 | 293.15 | |
| L.S.D at 0.05 | 22.86 | 11.35 | N.S. | N.S. | N.S. | 6.84 | N.S. | N.S. | |
| b) The effect of rootstocks | | | | | | | | | |
| Control | 109.53 | 111.46 | 21.96 | 24.58 | 42.43 | 35.87 | 294.78 | 295.35 | |
| L.g | 176.04 | 134.64 | 31.92 | 30.58 | 38.87 | 31.55 | 271.88 | 293.09 | |
| C.f | 132.54 | 105.75 | 27.38 | 26.25 | 47.96 | 41.42 | 292.40 | 447.87 | |
| C.m | 215.99 | 187.77 | 29.67 | 31.38 | 52.49 | 42.56 | 322.91 | 312.95 | |
| L.S.D at 0.05 | 24.39 | 25.32 | 2.79 | 4.09 | 3.86 | 5.88 | 13.37 | N.S. | |
| c) The effects of interaction between squash hybrids and rootstocks | | | | | | | | | |
| Control | 113.86 | 107.9 | 22.83 | 24.50 | 45.45 | 36.17 | 308.67 | 305.74 | |
| L.g | 158.42 | 148.5 | 32.25 | 32.17 | 42.38 | 36.89 | 288.67 | 299.52 | |
| Aziad | C.f | 141.46 | 122.0 | 31.25 | 27.33 | 50.02 | 46.08 | 274.85 | 192.43 |
| | C.m | 284.42 | 252.1 | 36 | 36.08 | 56.30 | 46.09 | 336.33 | 322.24 |
| Control | 105.19 | 115.0 | 21.08 | 24.67 | 39.40 | 35.57 | 280.89 | 284.96 | |
| L.g | 193.66 | 120.8 | 31.58 | 29 | 35.35 | 26.22 | 255.10 | 286.65 | |
| Raad | C.f | 123.62 | 89.5 | 23.50 | 25.17 | 45.90 | 36.75 | 309.95 | 297.31 |
| | C.m | 147.56 | 123.5 | 23.33 | 26.67 | 48.68 | 39.03 | 309.49 | 303.65 |
| L.S.D at 0.05 | 40.16 | 35.91 | 3.95 | N.S. | N.S. | N.S. | 18.91 | N.S. | |

Cont. = Self-grafted squash.

L.g = *Lagenaria siceraria*.

C.f = *Cucurbita ficifolia*.

C.m = *Cucurbita moschata*.

The effect of rootstocks

It was clearly that from data shown in Table (2) using *Cucurbita moschata* (C.m) as a rootstock resulted in significant increases in plant height compared to other rootstocks and control plants in both seasons. While, the highest number of leaves per plant was recorded when used the rootstock *Lagenaria siceraria* (L.g) followed by *Cucurbita moschata* (C.m) and lowest value was recorded with self-grafted plants in first season, while *Cucurbita moschata* (C.m) gave the highest value in second season. Concerning leaves dry weight, the obtained results showed that the highest value was recorded with *Cucurbita moschata* (C.m) followed by *Cucurbita ficifolia* (C.f) and *Lagenaria siceraria* (L.g), respectively, the lowest value were recorded with control treatment (self-grafted plants). The highest value of leaf area was recorded when used *Cucurbita moschata* (C.m) compared to the control in the first season only. But there were no significance differences detected among used different rootstocks and un-grafted plants in the second season.

The effect of interaction between squash hybrids and rootstocks

Effect of the interaction between squash hybrids and rootstocks shown in Table (2). Using Aziad hybrid as scion and *Cucurbita moschata* (C.m) as a rootstock led to a significant increase in plant height (cm) in both seasons compared to the rest treatments.

The results are shown that grafted Aziad on *Cucurbita moschata* (C.m) gives the highest increases in leaves number per plant and this increases was significantly difference in first season. The lowest value of leaves number per plant was recorded with self-grafted plants for both hybrids in both seasons. The results also showed that there were no significant differences when grafted both scions on different rootstocks and control on leaves dry weight in the first and second seasons. In addition, the highest value was recorded when grafted Aziad on *Cucurbita moschata* (C.m) in the first season only, but no significance differences noticed among different treatments in the second season. These results were agree with those obtained by Fouad et al. (2012) who pointed that the length of main stem, number of lateral stems and number of leaves of watermelon cv. Aswan F1 were significantly higher by grafting on 'Nun 6001 F1' in the first experiment. However, plants grafted

on 'Tetsukabuto F1' had the highest values in the second experiment in comparison to grafted plants onto other rootstocks and non-grafted plants. Also the results agree with Bekhradi et al. (2011) they reported that the highest values of survival rate of watermelon, stem length, number of lateral branches, number of internodes and fresh and dry weights, were recorded among grafted and control plants. Owing to using genetically different rootstocks all the studied characteristics were significantly affected by type of rootstocks used for grafting. El-Sayed et al. (2014) who indicated that the effect of grafting using different rootstocks on cucumber (*Cucumis sativus* L.) cultivar "Hady" in winter season caused a significant increase in plant height over self-grafted control plants. The results indicated that Ferro rootstock increased plant height of cucumber in summer and winter seasons as compared with un-grafted control. Alaaeldin et al. (2019) who indicated that grafting cucumber scion on Hersh rootstock showed a significance increase in plant length during first season, but the result didn't reach to the significant level in the second season in comparison to the control plants in both seasons, respectively. Star rootstock gave the highest value of leaves number per plant compared with the lowest number obtained by self-grafted control in both seasons, respectively. Also he pointed out that the highest significant leaf area value was achieved from Botle gourd rootstock in compared with Luffa rootstock which recorded the lowest value during both seasons.

The effect of squash hybrids

As shown in Table (3), the number of marketable fruits per plant showed no significant differences between both squash hybrids (Aziad and Raad) in the first season only, while, Aziad gave a higher significant value as scion than Raad in the second season. Also, the high values of marketable fruit weight/plant were given with using Aziad hybrid compared with Raad hybrid in both seasons. There are no significant differences noticed on average fruit weight with both hybrids used (Raad and Aziad) in the first season. While used Raad as the scion in the second season gave higher results for average fruit weight. In first season there were no significance differences between both hybrids (Aziad and Raad) for dry weight of 100 g of fresh fruit weight. While, outperformed Aziad with clear significance difference compared to Raad in the second season.

TABLE 3. The effect of different rootstocks on number of marketable fruits/plant, marketable fruit weight/plant, average fruit weight and dry weight of 100 g fruit fresh weight of squash plants grown under low plastic tunnel in winter season during seasons of 2015/2016 and 2016/2017.

| Measurements | Number of marketable fruits/plant | | Marketable fruit weight/plant | | Average fruit weight (g) | | Dry weight of 100 g fruit fresh weight (g) | | |
|--|-----------------------------------|------------------------|-------------------------------|------------------------|--------------------------|------------------------|--|------------------------|------|
| | 1 st season | 2 nd season | 1 st season | 2 nd season | 1 st season | 2 nd season | 1 st season | 2 nd season | |
| a) The effect of squash hybrids | | | | | | | | | |
| Aziad | 5.71 | 5.01 | 354.35 | 321.61 | 62.32 | 64.17 | 7.68 | 8.06 | |
| Raad | 5.08 | 4.39 | 321.52 | 288.11 | 63.81 | 66.09 | 7.39 | 6.01 | |
| L.S.D at 0.05 | N.S. | 0.29 | 11.66 | 18.02 | N.S. | 0.53 | N.S. | 1.10 | |
| b) The effect of rootstocks | | | | | | | | | |
| Control | 4.91 | 4.43 | 314.20 | 288.63 | 63.99 | 65.45 | 7.93 | 6.60 | |
| L.g | 3.77 | 3.55 | 248.04 | 242.54 | 65.75 | 68.40 | 5 | 6.96 | |
| C.f | 5.49 | 4.89 | 338.50 | 305.35 | 61.61 | 62.47 | 8.10 | 6.78 | |
| C.m | 7.41 | 5.95 | 450.99 | 382.92 | 60.90 | 64.21 | 9.11 | 7.82 | |
| L.S.D at 0.05 | 0.35 | 0.33 | 28.09 | 17.45 | N.S. | 2.38 | 2.12 | N.S. | |
| c) The effects of interaction between squash hybrids and rootstocks | | | | | | | | | |
| Control | 5.23 | 4.72 | 330.76 | 293.64 | 63.13 | 62.22 | 9.30 | 8.04 | |
| L.g | 3.83 | 3.55 | 244.88 | 235.62 | 63.94 | 66.27 | 4.80 | 7.32 | |
| Aziad | C.f | 5.45 | 4.85 | 331.67 | 304.13 | 60.87 | 62.70 | 8.30 | 8.18 |
| | C.m | 8.34 | 6.92 | 510.10 | 453.04 | 61.33 | 65.49 | 8.33 | 8.71 |
| Control | | 4.59 | 4.13 | 297.65 | 283.61 | 64.85 | 68.68 | 6.57 | 5.15 |
| | L.g | 3.72 | 3.54 | 251.20 | 249.46 | 67.57 | 70.53 | 5.20 | 6.60 |
| Raad | C.f | 5.54 | 4.93 | 345.34 | 306.57 | 62.34 | 62.24 | 7.90 | 5.37 |
| | C.m | 6.48 | 4.97 | 391.88 | 312.80 | 60.47 | 62.92 | 9.90 | 6.93 |
| L.S.D at 0.05 | | 0.49 | 0.46 | 39.72 | 24.68 | N.S. | 3.36 | N.S. | N.S. |

Cont. = Self-grafted squash.

L.g = *Lagenaria siceraria*.C.f= *Cucurbita ficifolia*.C.m= *Cucurbita moschata*.*The effect of rootstocks*

From the data in Table (3), using the rootstock *Cucurbita moschata* (C.m) had a significant increase of number of marketable fruits per plant compared to the others rootstock and self-grafted squash in the both seasons of study. Also, used rootstock *Cucurbita moschata* (C.m) gave the highest value of marketable fruit weight per plant compared to the lowest value that obtained by *Lagenaria siceraria* (L.g) in both seasons. On the other hand, there was no-significance difference between among

rootstocks and control in the first season only. However, in second season the highest value of the average fruit weight obtained by *Lagenaria siceraria* (L.g). The greatest results of dry weight of 100 g fruit fresh weight recorded with *Cucurbita moschata* (C.m) rootstock followed by *Cucurbita ficifolia* (C.f) then by the control. Whereas, the lowest value recorded by *Lagenaria siceraria* (L.g) in first season. There were no-significance differences among rootstocks genotypes in the second season on dry weight of 100 g fruit fresh weight.

The effect of interaction between squash hybrids and rootstocks

As for the interaction effect between hybrids and rootstocks, grafting squash hybrid Aziad onto *Cucurbita moschata* (C.m) rootstock had a significant increase on number of marketable fruits per plant compared to all treatments in the both seasons. As for, grafting (Aziad) on *Cucurbita moschata* (C.m) in the two seasons gave the highest value of marketable fruit weight per plant with clear significant differences compared to self-grafted plants. The average fruit weight parameter showed no significant difference between the grafted and self-grafted plants in first season only. While, the highest value of average fruit weight obtained by grafted Raad on *Lagenaria siceraria* (L.g) in the second season. Also the obtained results of dry weight of 100 g fruit fresh weight indicated that no significant differences between grafted and self-grafted plants in both seasons. These results are supported by the previous studies which revealed that grafted watermelon, using specific rootstocks increased plant growth, fruit yield, enhanced water transport and plant nutrition (Lee, 1994, Yetisir & Sari, 2003, Yetisir et al., 2007 and Han et al., 2009). Miguel et al. (2004) reported a similar effect of 'Shintoza' rootstock on fruit size in cultivar 'Queen' and 'Reina de Corazones'. In a study by Alaeldin et al. (2019) demonstrated that the highest commercial cucumber yield and total yield was revealed from Star rootstock and also, they indicated that the fruit number per plants showed significant differences between investigated treatments, the highest fruit number per plant was recorded in cucumber grafted on Star rootstock. Whereas, self-grafted plants (control) showed the significant decrease in fruit number per plant. Grafted cucumber scion on Star rootstock gave the highest fruits chemical contents in fruit dry matter % in both seasons.

The effect of squash hybrids

Data presented in Table (4) showed that there were no significant differences noticed when compared between squash fruits of Aziad and Raad concerning their TSS content in both seasons. However, Aziad showed the higher significant value of ascorbic acid content (V.C) compared to Raad in the first season only, but no significant difference between Aziad and Raad in the second season. Also a significant difference increase became clear of total carbohydrates when used Aziad hybrid compared to Raad hybrid in the first season only, while in second season

no significant difference detected between both hybrids Aziad and Raad.

The effect of rootstocks

Data shown in Table (4) revealed that TSS showed no significant differences among rootstocks genotypes in the first season. However, the highest significant value of total soluble solids (TSS) recorded by the self-grafted squash followed by *Cucurbita moschata* (C.m) in the second season. On the other hand, using *Cucurbita ficifolia* (C.f) gave the lowest value of Ascorbic acid content (V.C) compared with the control in the first season, while the highest value obtained by *Cucurbita ficifolia* (C.m) compared with the control in the second season. Moreover, the highest value of total carbohydrates (mg/g dry matter) occurred with using *Lagenaria siceraria* (L.g) as rootstock, but the lowest value of total carbohydrates occurred by *Cucurbita ficifolia* (C.f) in the first season without significant differences among all rootstocks and control plants.

The effect of interaction between squash hybrids and rootstocks

Data presented in Table (4) showed there were no significant differences between self-grafted plants of both squash hybrids and grafting on different rootstocks for TSS parameter in the first and the second seasons. The interaction between Aziad as a scion and *Cucurbita ficifolia* (C.m) as a rootstock gave the highest significant value of ascorbic acid content in the first season, but there are no significant differences between grafted and self-grafted plants in the second season. In the same regards, the interaction between Aziad as a scion and *Cucurbita ficifolia* (C.m) as a rootstock gave the highest significant value of total carbohydrates content compared to the lowest value obtained when Raad used as a scion and *Cucurbita ficifolia* (C.f) used as a rootstock in the first and second seasons. In the study of Paola et al. (2007) indicated that fruit dry matter, titratable acidity, total soluble solid contents, fruit firmness, and Hunter color (brightness, redness and yellowness) parameters of grafted melons were similar to those of the plants grown on their own roots. Obtained results agreed with the results for TSS%, ascorbic acid and total carbohydrate which have significant differences between investigated treatments, grafted cucumber scion on Star rootstock gave the highest value of TSS and ascorbic acid in both seasons. In contrast the lowest value of TSS and ascorbic acid were obtained from *Luffa* rootstock in both seasons.

While, Hersh rootstock reacted specifically to the total carbohydrates with the highest value compare to all treatments in both seasons. In contrary, the lowest total carbohydrates contents were found when grafted cucumber on Luffa followed by self-grafting cucumber in both seasons (Alaaeldin *et al.*, 2019). These findings disagreed with Villocino and Quevedo (2015) who stated that grafted watermelon (*Citrullus lanatus*) onto rootstocks of either squash Suprema (*Cucurbita maxima*) or bottle gourd Tambuli (*Lagenaria siceraria*) had much higher total soluble solids (TSS) content than those from ungrafted plants.

The effect of squash hybrids

Data presented in Table (5) revealed that fruits of squash hybrids Aziad and Raad showed no significant difference for content of calcium (Ca%) in both seasons. Regarding of total carotenoids the obtained results showed that there was non-significant difference between both hybrids in both seasons. In addition, the obtained results also concluded that free proline in squash fruits showed non-significant difference between Aziad and Raad hybrids in both seasons. Regarding, the results of leaf total chlorophyll Aziad hybrid gave a higher value than Raad in both seasons of study.

TABLE 4. The effect of different rootstocks on total soluble solids (TSS), ascorbic acid content (V.C) and total carbohydrates (mg/g dry matter) of squash plants grown under low plastic tunnel in winter season during seasons of 2015/2016 and 2016/2017.

| Measurements | Total soluble solids (TSS%) | | Ascorbic acid content (V.C) mg/100g juice | | Total carbohydrates (mg/g of dry matter) | | |
|--|-----------------------------|------------------------|---|------------------------|--|------------------------|-------|
| | 1 st season | 2 nd season | 1 st season | 2 nd season | 1 st season | 2 nd season | |
| a) The effect of squash hybrids | | | | | | | |
| Aziad | 6.14 | 5.11 | 24.29 | 23.19 | 33.98 | 35.79 | |
| Raad | 5.40 | 4.82 | 21.65 | 22.12 | 29.93 | 28.77 | |
| L.S.D at 0.05 | N.S. | N.S. | 1.45 | N.S. | 2.71 | N.S. | |
| b) The effect of rootstocks | | | | | | | |
| Control | 6.28 | 5.86 | 27.64 | 26.67 | 34.13 | 32.97 | |
| L.g | 5.60 | 4.25 | 19.09 | 18.20 | 35.16 | 34.68 | |
| C.f | 5.21 | 4.22 | 18.70 | 18.58 | 27.24 | 29.07 | |
| C.m | 5.99 | 5.54 | 26.47 | 27.16 | 31.30 | 32.4 | |
| L.S.D at 0.05 | N.S. | 0.55 | 1.34 | 3.08 | 3.28 | N.S. | |
| c) The effects of interaction between squash hybrids and rootstocks | | | | | | | |
| Aziad | Control | 7.77 | 5.77 | 29.12 | 28.30 | 31.47 | 34.28 |
| | L.g | 5.69 | 4.30 | 19.25 | 17.27 | 35.93 | 35.63 |
| | C.f | 5.28 | 4.54 | 19.27 | 18.88 | 31.27 | 35.81 |
| | C.m | 5.80 | 5.84 | 29.54 | 28.29 | 37.25 | 37.45 |
| Raad | Control | 4.80 | 5.94 | 26.16 | 25.04 | 36.78 | 31.67 |
| | L.g | 5.50 | 4.20 | 18.93 | 19.12 | 34.38 | 33.72 |
| | C.f | 5.13 | 3.90 | 18.13 | 18.27 | 23.21 | 22.33 |
| | C.m | 6.18 | 5.23 | 23.40 | 26.63 | 25.35 | 27.37 |
| L.S.D at 0.05 | N.S. | N.S. | 1.89 | N.S. | 4.64 | 6.61 | |

Cont. = Self-grafted squash.

L.g = *Lagenaria siceraria*.

C.f = *Cucurbita ficifolia*.

C.m = *Cucurbita moschata*.

TABLE 5. The effect of different rootstocks on Calcium, total carotenoids, free proline and leaf total chlorophyll of squash plants grown under low plastic tunnel in winter season during seasons of 2015/2016 and 2016/2017.

| Measurements | Calcium (Ca %) | | Total carotenoids (mg/g dry matter) | | Free proline (mg/g dry matter) | | Leaf total chlorophyll (SPAD) | | |
|--|------------------------|------------------------|-------------------------------------|------------------------|--------------------------------|------------------------|-------------------------------|------------------------|-------|
| | 1 st season | 2 nd season | 1 st season | 2 nd season | 1 st season | 2 nd season | 1 st season | 2 nd season | |
| a) The effect of squash hybrids | | | | | | | | | |
| Aziad | 0.693 | 0.771 | 0.31 | 0.24 | 0.23 | 0.24 | 72.66 | 71.94 | |
| Raad | 0.669 | 1.049 | 0.20 | 0.19 | 0.23 | 0.23 | 66.13 | 63.77 | |
| L.S.D at 0.05 | N.S. | N.S. | N.S. | N.S. | N.S. | N.S. | 2.59 | 4.89 | |
| b) The effect of rootstocks | | | | | | | | | |
| Control | 0.693 | 1.457 | 0.31 | 0.19 | 0.23 | 0.24 | 69.18 | 69.72 | |
| L.g | 0.657 | 0.67 | 0.20 | 0.17 | 0.18 | 0.18 | 60.85 | 61.65 | |
| C.f | 0.637 | 0.71 | 0.23 | 0.20 | 0.23 | 0.25 | 74.59 | 70.10 | |
| C.m | 0.737 | 0.803 | 0.30 | 0.28 | 0.27 | 0.27 | 72.96 | 69.95 | |
| L.S.D at 0.05 | N.S. | N.S. | N.S. | 0.08 | 0.01 | 0.02 | 2.59 | 2.47 | |
| c) The effects of interaction between squash hybrids and rootstocks | | | | | | | | | |
| Aziad | Control | 0.717 | 0.843 | 0.43 | 0.18 | 0.23 | 0.24 | 73.91 | 74.34 |
| | L.g | 0.737 | 0.72 | 0.21 | 0.20 | 0.19 | 0.19 | 59.25 | 60.43 |
| | C.f | 0.577 | 0.677 | 0.25 | 0.21 | 0.21 | 0.24 | 78.02 | 74.73 |
| | C.m | 0.74 | 0.843 | 0.37 | 0.37 | 0.28 | 0.27 | 79.46 | 78.26 |
| Raad | Control | 0.67 | 2.07 | 0.19 | 0.20 | 0.23 | 0.24 | 64.45 | 65.10 |
| | L.g | 0.577 | 0.62 | 0.19 | 0.14 | 0.18 | 0.18 | 62.45 | 62.88 |
| | C.f | 0.697 | 0.743 | 0.22 | 0.20 | 0.25 | 0.25 | 71.16 | 65.47 |
| | C.m | 0.733 | 0.763 | 0.22 | 0.20 | 0.26 | 0.26 | 66.45 | 61.63 |
| L.S.D at 0.05 | N.S. | N.S. | N.S. | N.S. | 0.02 | N.S. | 3.67 | 3.49 | |

Cont. = Self-grafted squash.

L.g = *Lagenaria siceraria*.

C.f= *Cucurbita ficifolia*.

C.m= *Cucurbita moschata*.

The effect of rootstocks

Data shown in Table (5) showed that all treatments had no significant differences on calcium percentage (Ca%) compared to control (self-grafted plants) in both seasons. The same results were obtained on total carotenoids when compared with the others rootstocks no significant differences detected among different rootstocks in first season only, while in second season the best result obtained when *Cucurbita moschata* (C.m) used compared with other rootstocks and control. Also, the obtained results showed that

the highest value of free proline was obtained by *Cucurbita moschata* (C.m) rootstock and the lowest value by grafting on rootstock *Lagenaria siceraria* (L.g) in both seasons. Moreover, the highest value of leaf total chlorophyll obtained by using *Cucurbita ficifolia* (C.f) as a rootstock compared with others and self-grafted plants in the first season. Also obtained results pointed out that there were a significant decrease in leaf total chlorophyll by using *Lagenaria siceraria* (L.g) compared with others and self-grafted plants in the second season.

The effect of interaction between squash hybrids and rootstocks

Data presented in Table (5) reported that the interaction effect between grafting of both scions of Aziad and Raad squash hybrids onto different rootstocks had no significant difference on calcium percentage (Ca%) and total carotenoids in the first and second seasons. The results of the interaction effect between all treatments in the first season recorded that the highest values of free proline and leaf total chlorophyll when grafted Aziad squash hybrid onto *Cucurbita moschata* (C.m) and the lowest values of free proline obtained with grafted Raad on *Lagenaria siceraria* (L.g), and for leaf total chlorophyll with grafted Aziad on *Lagenaria siceraria* (L.g). These findings were true in both seasons of the study. The free proline content showed no significant difference among all treatments and control in second season only. Similarly to the results obtained in the present study, Sun *et al.* (2010) who found that the calcium content of watermelon leaves was higher in grafted plants than in the non-grafted ones (control). Also, proline increase in the seedlings of 'Shangdong Thom' cucumber were grafted onto *Cucurbita ficifolia* under different low temperature stress conditions (Wang *et al.*, 1996).

(Stegemann and Bock, 2009) suggested that gene transfer is restricted to the contact zone between stock and scion indicates that the changes can become heritable only via lateral shoot formation from the graft site. Also Savvas *et al.* (2010) suggested that the uptake and/or utilization efficiency of macronutrients by plants may be enhanced by grafting onto some rootstocks.

These results may be due to the compatibility of grafting among rootstocks and cucurbita scion which improve absorption of water and elements and improved nutrient uptake in grafted plants that increases photosynthesis, which is particularly noticeable under less than optimal growing conditions such as weak sun light and low CO₂ content in greenhouses during winter condition. It has been suggested that these conditions allow grafted plants to produce higher yields, sometimes with improved fruit quality (Pulgar *et al.*, 2000, Xu *et al.*, 2006, Zhu *et al.*, 2006 and Hu *et al.*, 2006). Finally, we still need to know more about the grafting mechanism between rootstock and scion to confirm the changes genetically in long distance by using homogenous plants. The currently available tools are expected to advance our understanding resolve the long standing grafting mysteries.

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Conclusion

Under the condition of the experiment the benefit of growing squash crop under plastic low tunnel systems which applied in recent years worldwide (that allows a more efficient control of plants under different climate conditions). The obtained Results of this experiment highlighted the importance of checking the grafting compatibility with the most important hybrid used and concluded that grafting squash hybrid "Aziad" onto rootstock *Cucurbita moschata* (C.m) gives the highest yield and quality and reflects best solution to resistance to low temperature in winter season. This positive results may be due to the vigorous growth resulted of rootstock. Using of grafting technology enables those interested in the agricultural field to resistant different stress conditions such as low temperature stress, as the study showed.

Further work should be done on the wide range of rootstock under different climate condition in Egypt to find out the best one should be used for best productivity and quality, which has a good effect in reducing costs.

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Conflict of interest

Authors declare that there were no conflicts of interest.

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تأثير التطعيم على نباتات الكوسة المنزرعة تحت الأنفاق البلاستيكية خلال الموسم الشتوي

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أجريت تجربتين خلال موسمين متتاليين هما (2016-2015 / 2017-2016) بمحطة بحوث قها التابع لمركز البحوث الزراعي وتهدف التجربة إلى دراسة تأثير استخدام الأصول المختلفة للتطعيم على نباتات الكوسة لبعض صفات النمو الخضري: طول النبات (سم)، عدد الأوراق / نبات، مساحة الورقة (سم²)، الوزن الجاف للأوراق (جم) بالإضافة إلى النسبة الكلية للكلوروفيل (SPAD)، الكالسيوم (%)، الكاروتينات الكلية (مجم/جم مادة جافة)، البرولين (مجم/جم مادة جافة) والمحصول: عدد التمار التسويقية/ نبات، وزن التمار التسويقية/ نبات (جم)، متوسط وزن التمار (جم) وجودة التمار (الوزن الجاف/ 100 جرام وزن طازج تمار، المواد الصلبة الذاتية، حمض الاسكوريك، الكربوهيدرات الكلية (مجم/جم مادة جافة)). باستخدام هجين "رعد" وهجين "أزياد"، واستخدام 3 أصول هي: اليقطين (*Lagenaria siceraria* Standl)، (*Cucurbita ficifolia*)، الفرع الحسلي (*Cucurbita moschata*) بالإضافة إلى الكنترول وتم تنفيذ التجربة تحت الأنفاق البلاستيكية في الموسم الشتوي. وأشارت النتائج المتحصل عليها إلى أن أعلى قيمة لكلا من: طول النبات، عدد الأوراق/نبات، مساحة الورقة، النسبة الكلية للكلوروفيل، الكاروتينات الكلية، البرولين، حمض الاسكوريك عدد التمار التسويقية/نبات، وزن التمار التسويقية/نبات تم الحصول عليها عند تطعيم هجين الكوسة أزياد على أصل الفرع الحسلي *Cucurbita moschata* في كلا الموسمين. أما بالنسبة لصفات الوزن الجاف للأوراق، متوسط وزن التمار، الوزن الجاف/ 100 جرام وزن طازج تمار، الكالسيوم (%)، المواد الصلبة الذاتية، الكاروتينات الكلية فلم تظهر أي فروق معنوية واضحة بين تطعيم أيا من الهجينين (أزياد ورعد) على الأصول المختلفة مقارنة بالنباتات غير المطعومة.

وفي النهاية يمكن القول أن استخدام هجين أزياد كطعم على أصل الفرع الحسلي *Cucurbita moschata* تحت النفق البلاستيكي أعطى أعلى القيم للمحصول وجودة تماره مقارنة بالتطعيم على الأصول الأخرى ومقارنة بالنباتات غير المطعومة. مما يكون له تأثير جيد في خفض تكاليف الإنتاج وتعميم الربحية.