Investigation the heterosis, pollen and maternal tissue effects in qualitative characteristics of fruit in tomato

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Abstract
To investigate the magnitude of heterosis, metaxenia and cytoplasmic effects of qualitative traits, such as the percentage of total soluble solids, the percentage of titratable acidity, pH, total soluble solids/titratable acidity ratio, vitamin C, lycopene, β-carotene, total phenol, total flavonoids and antioxidant content of tomato fruit, five diverse tomato genotypes were evaluated in a complete diallel cross design. The analysis of variance showed that there is a high significant difference among lines and their hybrids for all the traits. There was a significant difference between hybrids especially among parents for all the traits due to metaxenia effect. As a result of cytoplasmic effect, highly significant differences were observed among some hybrids and their reciprocal crosses for all the traits. High amount of heterosis based on mid parents was observed for lycopene (157.92%) and vitamin C (100.00%), while high amount of heterosis based on the best parent was observed for lycopene (116.62%). The results are evidence for the existence of metaxenia, i.e. an effect of pollen on maternal tissues, in tomato fruits. The genetic variations in the different source of pollen can serve as the basis for selection of pollinizer to improve fruit quality depending on the market demand.

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Introduction
The cultivated tomato (*Lycopersicon esculentum* Mill.) is one of the most important vegetables and it contains a multitude of vitamins A, C, essential mineral and antioxidant properties which reduces particularly cancer types (Ray et al., 2011). For this reason, the production and consumption of tomato have increased in the recent years. Tomato which belongs to Solanaceae family is one of the most important vegetable crops in the worldwide (Benton, 2007).

Plant genetic resources are the reservoir of genetic recombination of genes and their variants, resulting from the evolution of plant species over centuries. Plant populations are the result of changes accumulated over time and concomitantly exposed the environment to the encountered.

Morphological parameters, as well as biochemical parameters, have been widely used in the evaluation of various crops (Rick and Holle, 1990; Kaemmerer et al., 1995). Exploitation of such traits increases our understanding of genetic variability available which could further facilitates their use in breeding for wider geographic adaptability with respect to biotic and abiotic stresses as well as short and long term breeding endeavors. The improvement program of tomato can be enhanced to a considerable extent if some basic information relevant to the pattern and magnitude of variability is made available to tomato breeders (Gulet et al., 2010).

Heterosis in tomato was first observed by Hedrick and Booth (1968) for higher yield and more number of fruits per plant. Choudhary et al. (1965) emphasized the extensive utilization of heterosis to step up tomato production. Heterosis term in tomato is in the form of the greater vigor, faster growth and development, earliness in maturity, increased productivity, higher levels of resistance to biotic and abiotic stresses (Yordanov, 1983), while Gulet et al. (2010) indicated highly heterosis for fruit length, fruit diameter and fruit weight.

The pollen from different sources affects readily discernible characteristics of seeds and fruits in the period immediately following fertilization have been noted for well over a century (Denney, 1992). These effects on tissues of purely maternal origin, rather than on parts resulting from syngamy, have also been termed “metaxenia” (Swingle, 1928). In fleshy fruits, such as date, apple and persimmon, it is found that “metaxenia” expressed in the maternal tissues of carpels and accessory tissue (Denney, 1992). Some metaxenia effects were attributed to seed-controlled hormonal level (Mizrahi et al., 2004). Although the principal cause of metaxenia is not unambiguously understood, this phenomenon may be put to immediate use in horticultural practice (Mizrahi et al., 2004).

The simplest and most probable theory to explain metaxenia is that the embryo or endosperm, or both of them, secrete hormones, or soluble substances analogous to them, which diffuse out into the tissues of the mother plant that constitute the seed and fruit and there exert a specific effect on these tissues, varying according to the particular male parent used to fecundate the embryo and endosperm (Swingle, 1928). Metaxenia is the effect of pollen on fruit shape and other fruit characteristics. Metaxenia may be able to be used to identify the best pollinizer parents to decrease fruit development period and increase yield in mixed cultivar plantings (Olfati et al., 2010). Metaxenia, the transmission of traits from the pollinizer to the female’s tissues, is a phenomenon hitherto unknown in tomatoes (Piotto et al., 2013).

In 1979, Freytag reported “metaxenia” effects on the development of pod size in common bean. He observed normal maternal control of pod development in bean, was influenced by rate of elongation and total length of the mature pod when flowers were fertilized by foreign pollen. Olfati et al., 2010 demonstrated metaxenia on cucumber fruit characteristics. They indicated depending on the intended purpose of cucumber production, appropriate parental lines will be different. Piotto et
Rahaii et al. (2013) investigated metaxenia effects on pilose fruit surface of *Solanumlycopersicum*. The pollen of *S. galapagense* was able to raise pilosity in the crossed fruits.

Metaxenia and xenia have been mainly proposed for improving maize yield (Weingartner et al., 2002) and several fruit crops, such as pecan nuts, pistachio nuts and avocado (Robbertse et al., 1996; Sedgley and Griffin, 1989), but especially date palms (Nixon, 1928; Shaheen et al., 1989). In contrast, to our knowledge, no instances of this phenomenon have ever been described in morphological and biochemical traits of fruit in tomato. Previous studies have reported that pollen had an effect on maternal tissue characteristics in some crops (Denney, 1992; Wallace and Lee, 1999; Mizrahi et al., 2004). Althought tomatoes are self-fertile but, as well as other members of the Solanaceae require a special kind of pollinator to achieve proper fruit set. Today the growers use bumblebee as pollinator because of the lower production costs, increased yields, and improved fruit quality and this is caused cross pollination (Velthuis and van Doorn, 2006). The present study was therefore conducted to estimate the magnitude of heterosis and metaxenia and cytoplasm effects for qualitative traits in tomato fruit in crosses using five diverse tomato genotypes in complete diallel combinations.

**Materials and methods**

**Plant material**

Five lines of *Solanumlycopersicum*, named CLN1621F (A), CLN2463E (B), CLN2071D (C), CLN1462A (D) and CLN3126A-7 (E), which were received from 'The World Vegetable Center (AVRDC)', were used for this experiment. All lines varied for qualitative traits. These lines were grown in a randomized complete design with three replications, with a culture intensity of 3 plants/m². Hand pollinations were made in a greenhouse and the crossed flower was covered from unfavorable pollination. The lines were crossed in a complete diallel cross experiment with reciprocals. Ripe fruits were selected randomly to estimate quality traits.

**Fruit analysis**

Total Soluble Solids (TSS) is an index of soluble sugars content in fruit. Ground tomato tissues homogenate were filtered and TSS (ºBrix) of flowthrough was determined by a refractometer (CETI-BELGUM).

Titretable acidity was measured in aqueous pulp extracts. Five grams of pulp tissue were homogenized with 5 mL of double-distilled water with a mortar and pestle. 10 mL of double-distilled water was then added to the homogenates. After centrifugation (10000 g, 10 min) the pH of the supernatant was determined and titratable acidity was measured by titration with 0.1 N NaOH to pH 8.2.

TSS/TA was calculated by dividing the total soluble solids to titratable acidity.

Determination of ascorbic acid content was performed according to the method of Boret et al. (2006). Vegetable extracts (0.1 g) were extracted with 10 mL of 1% metaphosphoric acid. After the vegetable extracts were filtered, the filtrate (1 mL) was added to 9 mL of 50 μM 2,6-dichloroindophenol (DIP), and the absorbance at 515 nm was read with a spectrophotometer (PG Instrument + 80, Leicester, United Kingdom). The results were expressed as milligrams of AsA per 100 g of FW.

Flavonoid content was measured spectrophotometrically. A total of 1 mL of water vegetable extracts (200 μg/mL) was added to 5.7 mL of distilled water and 0.3 mL of 5% Sodium nitrite. After 5 min, 3 mL of 10% Aluminum chloride was added 5 minlater. After another 6 min, 2 mL of the mixture solution was added 102 mL of 1 N NaOH. Absorbance was measured at 510 nm using spectrophotometer. Catechin was used as the standard for a calibration curve (Boret et al., 2006).

The content of total phenolic compounds in vegetable
extracts was measured according to the method of Boret et al. (2006) and calculated using gallic acid as a standard. The vegetable extract (0.1 g) was dissolved in 5 mL of 0.3% HCl in methanol/water (60:40, v/v). The resulting solution (100 μL) was added to 2.0 mL of 2% Sodium carbonate. After 2 min, 50% Folin-Ciocalteu reagent (100 μL) was added to the mixture, which was then left for 30 min. Absorbance was measured at 750 nm using a spectrophotometer.

b-Carotene and lycopene were determined according to the method of Navarro et al. (2006). Fresh samples of tomato fruit were homogenized using a pestle and mortar in the presence of liquid N2. 60 mL of acetone–hexane (4:6) solvent were added to 1.0 g of tomato homogenate and mixed in a test-tube. Automatically, two phases separated, and an aliquot was taken from the upper solution for measurement of optical density at 663, 645, 505, and 453 nm in a spectrophotometer. Lycopene and b-carotene contents were calculated according to below equations:

\[
\text{Lycopene (mg 100 ml}^{-1}\text{of extract)} = -0.0458 \times A_{663} + 0.204 \times A_{645} + 0.372 \times A_{505} - 0.0806 \times A_{453}
\]

\[
\text{b–Caroten (mg 100 ml}^{-1}\text{of extract)} = 0.216 \times A_{663} - 1.22 \times A_{645} - 0.304 \times A_{505} + 0.452 \times A_{453}
\]

Lycopene and b-carotene were finally expressed as mg/100g fW.

The antioxidant activity was expressed as the percentage of decline of the absorbance, relative to the control, corresponding to the percentage of DPPH that was scavenged.

The percentage of DPPH, which was scavenged (%DPPHsc), was calculated using:

\[
\%\text{DPPHsc} = \frac{A_{\text{cont}} - A_{\text{samp}}}{A_{\text{cont}}} \times 100
\]

Where \(A_{\text{cont}}\) is the absorbance of the control, and \(A_{\text{samp}}\) the absorbance of the sample.

Statistics

Analysis of variance, according to randomized complete design, was carried out to evaluate the significant differences between lines and hybrids using the general liner model (GLM) procedure of SAS software ver. 9.1 (SAS Institute, 2003). Comparison of means was performed for evaluation of metaxenia and cytoplasmic effects using the LSD test at 5% significance probability level.

Heterosis for all studied traits was calculated based on the best parent heterosis (Hbp) and mid-parents heterosis (Hmp) approach. Mid-parent heterosis (MPH) was calculated in terms of percent increase (+) or decrease (-) of the F1 hybrids against its mean parent value as suggested by Fehr (1987).

\[
\text{MPH} (%) = \left[\frac{\text{F1} - \text{MP}}{\text{MP}}\right] \times 100
\]

Similarly, the best parent heterosis (BPH) was also estimated in terms of percent increase or decrease of the F1 hybrid over its the best parent.

\[
\text{BPH} (%) = \left[\frac{\text{F1} - \text{BP}}{\text{BP}}\right] \times 100
\]

Results

The analysis of variance showed high significant differences \((p<0.01)\) among lines and their hybrids for all the studied traits (Table 1). Comparison of means by LSD method \((p<0.05)\) for all traits are presented in Table 2. There was a significant
difference between generations especially among parents for almost measured traits. Mid-parent heterosis and the best parent heterosis were differently ranged in hybrids for all studied traits (Table 3 and 4).

**Antioxidant content (Inhibition of DPPH%)**
The comparison of means for antioxidant content showed that there is a significant difference in D×B hybrid. The lowest amount of antioxidant content was observed in D×B hybrid (36.15% Inhibition of DPPH). On the other hand, D×B and its reciprocal cross were significantly different at %5 level which was due to pollen and cytoplasmic effects on the value of antioxidant content. Mid-parent heterosis ranged from -53.09 to 2.51% and the best parent heterosis was ranged from -53.46 to 1.70% (Table 3 and 4).

Highly negative heterosis was observed for hybrid D×B, rather than mid parent and the best parent.

### Table 1. Analysis of variance between various genotypes for all studied traits in tomato.

<table>
<thead>
<tr>
<th>Source of variable</th>
<th>Degree of freedom</th>
<th>Mean of squares</th>
</tr>
</thead>
<tbody>
<tr>
<td>line and their hybrids</td>
<td>24</td>
<td>Y1 388.71** Y2 7693.11** Y3 461.29** Y4 48.55** Y5 9.64** Y6 6.70** Y7 0.32** Y8 5.98** Y9 2.52** Y10 0.07**</td>
</tr>
<tr>
<td>Error</td>
<td>50</td>
<td>6.54 5.26 4.49 0.83 0.03 0.02 0.00 0.02 0.00</td>
</tr>
<tr>
<td>CV %</td>
<td>-</td>
<td>3.47 3.43 7.79 8.55 4.72 7.32 0.95 3.59 2.36 4.30</td>
</tr>
</tbody>
</table>

** Significant at 1% probability level.

Y1: antioxidant content (%Inhibition of DPPH), Y2: Total Flavonoid (mg Catechin/100gFW), Y3: Total Phenol (mg Galic acid/100gFW), Y4: Vitamin C (mg AsA/100gFW), Y5: Lycopene (mg/100gFW), Y6: β-carotene (mg/100gFW), Y7: pH, Y8: TA, Y9: TSS%, Y10: TSS/TA.

**Total Flavonoid (mg Catechin/100gFW)**
The results of ANOVA indicated that there is a significant difference among all studied genotypes for total flavonoid (Table 1). The A line had the highest amount of total flavonoid content (176.76 mg Catechin/100gFW) while lowest amount of total flavonoid content (26.48 mg Catechin/100gFW) was observed in line D. Different Source of pollen also caused significantly several variations in the fruit total flavonoid content between A female line and its' hybrids, however the difference between A×C and A×D was not significant (Table 2).
| LSD (P=0.05) | 4.195 | 3.762 | 3.356 | 1.492 | 0.262 | 0.244 | 0.066 | 0.401 | 0.247 | 0.068 |

Y1: antioxidant content (%Inhibition of DPPH), Y2: Total Flavonoid (mg Catechin/100gFW), Y3: Total Phenol (mg Galic acid/100gFW), Y4: Vitamin C (mg AsA/100gFW), Y5: Lycopene (mg/100gFW), Y6: β-carotene (mg/100gFW), Y7: pH, Y8: TA, Y9: TSS%, Y10: TSS/TA.
The highest total flavonoid content was observed when A line was used as maternal parent (Table 2). It may be as a result of expression of mid-parent heterosis for hybrids with A line as common parent, but the best parent heterosis was negative for these hybrids. The result showed that crossing of A line with different pollen samples and their reciprocal cross can cause significantly variations total flavonoid content of fruit (Table 2). It could be concluded metaxenia and cytoplasmic effects controls the total flavonoid content of fruit.

Table 3. Extent of mid-parent heterosis (%) for all studied traits in 25 hybrids of tomato from an 5×5 complete diallel.

<table>
<thead>
<tr>
<th>Female</th>
<th>Male</th>
<th>Y1</th>
<th>Y2</th>
<th>Y3</th>
<th>Y4</th>
<th>Y5</th>
<th>Y6</th>
<th>Y7</th>
<th>Y8</th>
<th>Y9</th>
<th>Y10</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>B</td>
<td>1.35</td>
<td>42.66</td>
<td>52.74</td>
<td>20.80</td>
<td>12.07</td>
<td>-14.51</td>
<td>0.49</td>
<td>13.65</td>
<td>17.36</td>
<td>4.65</td>
</tr>
<tr>
<td>A</td>
<td>C</td>
<td>-1.69</td>
<td>52.59</td>
<td>-3.01</td>
<td>-37.73</td>
<td>157.92</td>
<td>-24.92</td>
<td>-5.32</td>
<td>27.70</td>
<td>12.79</td>
<td>-11.52</td>
</tr>
<tr>
<td>A</td>
<td>D</td>
<td>0.97</td>
<td>57.55</td>
<td>47.64</td>
<td>4.80</td>
<td>20.59</td>
<td>-4.81</td>
<td>-7.78</td>
<td>28.48</td>
<td>33.47</td>
<td>0.53</td>
</tr>
<tr>
<td>A</td>
<td>E</td>
<td>2.41</td>
<td>41.10</td>
<td>92.72</td>
<td>95.34</td>
<td>10.17</td>
<td>-16.74</td>
<td>-3.76</td>
<td>-12.98</td>
<td>6.61</td>
<td>19.18</td>
</tr>
<tr>
<td>B</td>
<td>A</td>
<td>1.77</td>
<td>-53.85</td>
<td>-32.73</td>
<td>35.13</td>
<td>27.32</td>
<td>9.39</td>
<td>-1.98</td>
<td>7.16</td>
<td>11.77</td>
<td>5.81</td>
</tr>
<tr>
<td>B</td>
<td>C</td>
<td>-1.15</td>
<td>-52.64</td>
<td>-26.27</td>
<td>-52.04</td>
<td>-18.72</td>
<td>20.00</td>
<td>-2.96</td>
<td>32.99</td>
<td>11.26</td>
<td>-16.28</td>
</tr>
<tr>
<td>B</td>
<td>D</td>
<td>1.19</td>
<td>50.24</td>
<td>44.29</td>
<td>56.22</td>
<td>25.00</td>
<td>41.28</td>
<td>-10.14</td>
<td>38.72</td>
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</tr>
<tr>
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<td>7.16</td>
<td>0.82</td>
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<td>-3.54</td>
<td>2.94</td>
<td>17.36</td>
<td>11.63</td>
</tr>
<tr>
<td>C</td>
<td>A</td>
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<td>-61.90</td>
<td>-29.15</td>
<td>-26.53</td>
<td>-62.90</td>
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<td>5.32</td>
<td>17.05</td>
<td>29.35</td>
<td>10.30</td>
</tr>
<tr>
<td>C</td>
<td>B</td>
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<td>-23.31</td>
<td>-18.72</td>
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<td>C</td>
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<td>E</td>
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<td>24.54</td>
<td>53.17</td>
<td>60.74</td>
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<td>25.52</td>
<td>4.93</td>
<td>9.01</td>
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<td>18.79</td>
</tr>
<tr>
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<td>-67.86</td>
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<td>-10.95</td>
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<tr>
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<td>57.22</td>
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<td>119.51</td>
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<td>-4.32</td>
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<td>-1.67</td>
<td>17.65</td>
<td>3.87</td>
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<td>14.09</td>
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<td>-5.55</td>
<td>21.08</td>
<td>18.13</td>
<td>-9.09</td>
</tr>
</tbody>
</table>

Y1: antioxidant content (%Inhibition of DPPH), Y2: Total Flavonoid (mg Catechin/100gFW), Y3: Total Phenol (mg Galic acid/100gFW), Y4: Vitamin C (mg AsA/100gFW), Y5: Lycopene (mg/100gFW), Y6: β-carotene (mg/100gFW), Y7: pH, Y8: TA, Y9: TSS%, Y10: TSS/TA.

The different source of pollen also caused several variations in the total flavonoid content of fruit in B line’s hybrids. The highest total flavonoid content of fruit was observed in B×D hybrid (63.95 mg Catechin/100gFW). Other sources of parental pollen which crossed with B line had significant differences with each other and their reciprocals in total flavonoid content of fruit. It could be the result of metaxenia and cytoplasmic effects on the total flavonoid content of fruit. The highest percent of mid-parent and the best parent heterosis was observed in the crosses between B line and D pollen (50.24 and 9.04 mg Catechin/100gFW, respectively). Different sources of pollen also caused several variations in the total flavonoid content in C line. Among the pollen, the D pollen showed highest total flavonoid content of fruit (55.01 mg Catechin/100gFW) in C line which followed by C×E hybrid (47.04 mg Catechin/100gFW). Other pollen samples which crossed with C line showed significant differences with each other.
and their reciprocal crosses in total flavonoid content of fruit. It could be a result of metaxenia and cytoplasmic effects on controlling the total flavonoid content of fruit. Among studied lines, the D line had the lowest of total flavonoid content (26.48 mg Catechin/100gFW), also parental pollen crossed with D line as a maternal showed significant difference with their reciprocals. The difference between hybrids and their reciprocals for cross of D and E lines with other sources of pollen was significant. It was indicated that cytoplasmic effect could control the total flavonoid of fruit. Also, different sources of pollen also caused several variations in the fruit total flavonoid of E line, which indicated metaxenia effect. The highest mid- parent and the best parent heterosis were observed in C×D hybrid (71.40% and 45.88%, respectively) for total Flavonoid content.

Table 4. Extent of the best parent heterosis (%) for all studied traits in 25 hybrids of tomato from an 5×5 complete diallel.

<table>
<thead>
<tr>
<th>Female</th>
<th>Male</th>
<th>Y1</th>
<th>Y2</th>
<th>Y3</th>
<th>Y4</th>
<th>Y5</th>
<th>Y6</th>
<th>Y7</th>
<th>Y8</th>
<th>Y9</th>
<th>Y10</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>B</td>
<td>0.75</td>
<td>-5.00</td>
<td>28.88</td>
<td>5.00</td>
<td>11.91</td>
<td>-22.69</td>
<td>-0.25</td>
<td>10.47</td>
<td>-0.57</td>
<td>-9.09</td>
</tr>
<tr>
<td>A</td>
<td>C</td>
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<td>-25.94</td>
<td>-49.54</td>
<td>58.33</td>
<td>-54.10</td>
<td>-10.31</td>
<td>27.51</td>
<td>1.66</td>
<td>-20.65</td>
</tr>
<tr>
<td>A</td>
<td>D</td>
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<td>-1.75</td>
<td>2.38</td>
<td>1.67</td>
<td>-13.16</td>
<td>-13.52</td>
<td>8.66</td>
<td>28.85</td>
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</tr>
<tr>
<td>A</td>
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<td>0.93</td>
<td>-14.35</td>
<td>30.83</td>
<td>25.69</td>
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Y1: antioxidant content (%Inhibition of DPPH), Y2: Total Flavonoid (mg Catechin/100gFW), Y3: Total Phenol (mg Gallic acid/100gFW), Y4: Vitamin C (mg AsA/100gFW), Y5: Lycopene (mg/100gFW), Y6: β-carotene (mg/100gFW), Y7: pH, Y8: TA, Y9: TSS%, Y10: TSS/TA.

Total Phenol (mg Gallic acid/100gFW)
There was a significant difference in the total phenol of fruit among all varieties. The different source of pollen also caused several variations in the total phenol of fruit in A line which could be an effect of metaxenia. The highest amount of total phenol of fruit was produced by E and B pollen in hybridization with A line (56.95 and 56.10 mg Gallic acid/100gFW, respectively) which could be a result of the best parent heterosis. The highest mid- parent and the best parent heterosis were recorded for A×E (92.72 and 30.83, respectively). Hybrids generated from A showed higher mid- parent heterosis except for A×C. The crosses of A lines with other lines pollen showed
significant differences with their reciprocals, which indicated that cytoplasmic effect could be effective in controlling the total phenol of fruit. The analysis of variance showed that there is a significant difference between B×A and B×E hybrids for the total phenol of fruit. The highest amount of total phenol was observed in B×D hybrid with 31.99 mg Gallic acid/100gFW which could be as a result of heterosis (44.29% and 6.88% for mid-parent and the best-parent, respectively). There was a significant difference among C parent, C×E and C×D hybrids for total phenol of fruit. In the cross of C as a female line with D and E pollens, it produced the highest amount of total phenol of fruit, while other crosses didn't show significant variations in the total phenol of fruit. In the crosses with the C line as a female parent, the highest amount of the total phenol of fruit was obtained when the pollen used from D and E lines. The other combination produced lower total phenol of fruits. The value of mid-parent heterosis was high in C×D and C×E hybrids (58.14 and 53.17, respectively). The comparison of reciprocals with hybrids which C lines was used as a maternal parent on them showed different result which could be as a result of cytoplasmic inheritance to controlling the phenol of fruit. The A and C strains of pollen also caused several variations in the total phenol of D maternal line. Among the hybrids, only D×A and D×C showed comparatively higher total phenol of fruit than other hybrids. When the pollen from B line was crossed with the E line, it produced significant differences in the total amount phenol but other strains of pollen did not cause variations in the total phenol of fruit in E line. The highest amount of total phenol was resulted in E×B hybrid (20.86 mg Gallic acid/100gFW).

The reciprocal crosses of all hybrids were significantly different at the 5% probability level which could be as a result of the cytoplasmic effects inheritance to controlling the Total Phenol of fruit. The crossing of E and D lines with all pollens showed negative the best parent heterosis in the total phenol of fruit. The highest value of mid-parent and the best parent heterosis was recorded for C×D hybrid (71.40% and 45.88%, respectively).

Vitamin C (mg AsA/100gFW)
There was a significant difference in the vitamin C content of fruit among all studied lines. The vitamin C content of A line showed significant variations when it was pollinated with different pollen strains. The pollination of A with E line produced the highest amount of vitamin C in fruit (16.34 mg AsA/100gFW) which could be a result of heterosis. The heterosis based on mid- parent and the better parent was estimated 95.34% and 25.69% respectively. Different strains of pollen also caused several variations in the vitamin C of fruit in B, C and D line. Also, the B×D and the B×A hybrids had the highest levels of the vitamin C in their fruit (17.20 and 15.27 mg AsA/100gFW, respectively). The mid-parent and the best parent heterosis were 56.22 and 35.13, respectively. The heterosis caused higher vitamin C content in C×E hybrid rather than their parents. The lowest vitamin C of fruit was observed in D×B hybrid and the highest one in D×C hybrid (6.90 and 16.69 mg AsA/100gFW, respectively). Also, the highest amount of mid-parent heterosis was estimated for vitamin C of fruit by D×E (100%) and A×E (95.34%) and E×C (73.43%) hybrids, respectively. The pollination of E line with different sources of pollen resulted in different content of vitamin C in hybrid's fruits, but their difference were not significant. The amount of vitamin C in E line was increased with pollination by all of pollen source but the lowest vitamin C of fruit was produced by the crosses between E line as female with D pollen. Highly significant differences were observed among A×B, A×E, B×D, B×E, C×D and D×E hybrids and their reciprocal crosses for vitamin C which was due to cytoplasmic effect.

Lycopene (mg/100gFW)
The results of the analysis of variance showed that there is a significant difference among lines was significant for lycopene content of fruit except for A and B lines. The pollens from A and B lines caused
several variations in the lycopene content of the A line fruit. The highest amount of lycopene in A lines were observed when it pollinated with C and B lines, respectively (5.70 and 4.04 mg/100g FW, respectively). In the crosses of A, C and E pollen with B line as female, there were several variations in the lycopene of fruit. The pollens from E and A lines produced the highest amount of lycopene of fruit, while other sources of pollen induced any significant variations in lycopene of fruit. The amount of the lycopene of fruit in C line and its hybrids pollinated with different sources of pollen was less than 1 mg/100gFW, their difference to each other. It is indicated that if C line with orang fruits was crossed as female parent with other pollens, lycopene level in fruit would decreased because of cytoplasmic effect. The amount of the lycopene of the fruit in D line was significantly increased when pollinated with varied pollens. Among the hybrids, the highest amount of lycopene of fruit was produced in D×B hybrid (7.82 mg/100gFW), while the lowest amount of lycopene was produced by D×C hybrid (2.47 mg/100gFW). The all hybrids showed significant differences with their reciprocals in lycopene content. The amount of the lycopene of fruit in E line was significantly increased when pollinated with the pollens of B and C lines. The pollens of B and C lines also caused variations in the lycopene content of fruit of E line, while other lines variations in the lycopene content of fruit. Heterosis varied from -65.35 to 157.92% over mid-parent. Maximum value of heterosis over mid parent was observed in A×C (157.92%) followed by D×B (157.24%), D×C (143.77%) and E×C (119.51%). The highest heterosis over the best parent was observed in D×A (116.62%) followed by D×C (62.35%) and A×C (58.33%).

β-carotene (mg/100gFW)

The result of analysis of variance showed that difference between lines was significant for content of β-carotene of fruit. The highest β-carotene content of fruit was produced in the C line (5.12 mg/100g FW) and lowest value was resulted with D (0.82 mg/100gFW). The cross of A line with pollen of C line produced the highest amount of β-carotene which significantly different from pollination with other sources of pollen. The amount of β-carotene of fruit in B line was significantly varied when pollinated with various pollens except C pollen. The B×E hybrid had the highest amount of β-carotene of fruit (5.01 mg/100gFW), while other pollen sources showed little variations in the β-carotene of fruit. The color of fruit in C line was orange and the highest amount of β-carotene was observed in this line (5.12 mg/100gFW). The cross of C line as a female with pollens of D and E decreased β-carotene content in fruit, while others source of pollen showed little decrease in the β-carotene content of fruit, which were significantly different from C line. The highest positive mid-parent heterosis was observed for two hybrids C×A (72.20%), C×B (73.05). The comparison of hybrids common in C line as female with reciprocal crosses showed significant differences in β-carotene content, it is indicated that cytoplasmic effect may control β-carotene content of fruit. The cross of D line with pollens of A and B lines increased the β-carotene content of fruit. The pollination of E line with C pollens significantly decreased β-carotene content while other source of pollens had not any significant impact on it. The cytoplasmic effect was significant between all hybrids and their reciprocals except between A×B and B×A hybrids. The highest mid-parent heterosis was recorded for C×B (73.05%) followed by C×A (72.20%). The estimates of heterosis over the best parent ranged from -76.45% for cross E×C to 26.54% for cross B×E (Table 4). Negative heterobeltiosis was found in often hybrids.

pH

The results of the analysis of variance indicated that there is a significant difference among most lines except between A and B line. The pollen of E line caused variations in the pH of A line fruit, but it’s difference with other hybrids was not significant (Table 2). The pH of fruit was significantly different for all hybrids with B mother in common expect for the cross with pollen derived from A line. The
different strains of pollen also did not cause any variations in the pH of fruit in the C line, except for D pollen. Also, all of pollens caused several variations in the pH of D maternal line. The cross of E female line with B pollen didn’t show any significant variations in pH of fruit. The cytoplasmic effect was significant between all hybrids and their reciprocal crosses. The mid-parent and the best parent heterosis were high hybrids with D parent in common. The highest mid-parent and the best parent heterosis were recorded for D×B (11.52 and 3.86%, respectively).

Titratable acidity%
There is a significant difference among lines for TA% of fruit. The TA% content of fruit in lines ranged from 4.55% to 6.97%. The pollination of A line with other lines pollen produced significantly different TA% content from A line, except for E line. The highest value of TA% of fruit was produced by A×C (8.39%), while the lowest amount produced by A×E (5.90%), which had not significant difference with A line. C and D pollens also caused several variations in TA% content of B line fruit, which was increased TA%. The highest TA% of fruit was produced in B×C hybrid (9.01%), but its difference from B line and other hybrids was not significant. The crosses of C line with pollen of other lines significantly increased the TA% of fruit. The A and E pollens produced significant differences in the TA% of fruit in D line, but other strains of pollen did not cause variations in TA% in D line. The highly amount of TA% of fruit was produced by C×D hybrid (7.89%). Also, highly positive mid parent heterosis was observed in C×D hybrid (42.03%). All pollens showed comparatively variation in TA% in E fruits, while D pollen didn’t show any significant alter in TA% of fruit. The highest value of TA% of fruit was produced in E×A hybrid (9.61%), while the lowest TA% was produced by E×C (5.58%). The cytoplasmic effect was significant between all hybrids and their reciprocal, with an exception between B×E and E×B hybrids. The mid-parent and the best parent heterosis was high in the hybrids of B female with other pollens. The highest mid-parent heterosis was recorded in C×D (42.03%) which followed by E×A (41.74%), B×D (37.72%) and B×C (32.99%), while the highest the best parent heterosis was observed for hybrids E×A (37.68%), B×C (29.27%) and A×C (27.51%), respectively.

Total soluble solids% (TSS %)
All of lines had significant differences in the TSS% of fruit except for A and E lines. All of pollens also caused several variations in the TSS% in A maternal line, which increased content of TSS in A hybrids. The A and C pollens also caused several variations in TSS% in B line fruit, but other pollens did not cause variations in the TSS% fruit in B line. All of pollens also caused several variations in the TSS% in C line fruit which increased content of TSS in C line. The B and E pollens also caused several variations in TSS% in D line fruit. The highest amount of TSS% was produced in D×E hybrid. Also the crosses of E line with other lines pollen showed significant differences in the TSS% of fruit. The highest amount of TSS% of fruit was produced in E×B (7.50%). The cytoplasmic effect between all hybrids and their reciprocal was significant except between B×C and C×B. The highest mid-parent heterosis was recorded in hybrids E×A (62.42%) followed by A×D (33.47%), C×D (36.60%) and C×E (33.76%), while the highest the best parent heterosis was observed in hybrids E×A (69.42%) for total soluble solids%. Bhatt et al., (2004) and Singh et al. (2008) also reported significant and high heterosis over the best parent in tomato for total soluble solids.

TSS/TA
There was a significant difference among all studied lines for TSS/TA except between A and E. All of pollens caused variation in TSS/TA content of A maternal line except of C pollen, which its ratio was lower than others. A, B, C and D pollens also caused variations in TSS/TA in B line, which its value was lower than one because of their sour taste. None of pollens also caused several variations in the TSS/TA in C line fruit. All of pollens caused variation in TSS/TA content of D line fruit, expect C pollen, while the value of TSS/TA was higher than one because of their sweet taste. The pollination of E line with other
source of pollen produced significant differences in the TSS/TA ratio of fruit for A pollen, and the TSS/TA ratio for E×B was obtained above one, it could be a reason of its sweet taste. While the value of TSS/TA was lower than one because of their sour taste. Highly significant differences were observed among A×C, A×D, B×C, B×D, B×E, C×D and D×E hybrids and their reciprocal crosses for TSS/TA of fruit in the result of cytoplasmic effect. Mid-parent heterosis ranged from -19.25 to 33.69% and the best parent heterosis ranged from -27.27 to 16.44% (Table 3 and 4). The highest mid-parent heterosis was recorded in hybrid D×A (33.69%) followed by D×E (32.62%), while the highest the best parent heterosis was observed for hybrids A×E (19.18%) and E×A (16.44%).

Discussion

The results show that the tomato fruits exhibit metaxenia, since the tissues affected by the pollen source are solely of maternal origin (Osman et al., 1974; Daulta and Chauhan, 1983, 1984; Kahn et al., 1994; Chaudhary and Dessay, 1995; Nerd and Mizrahi, 1997).

The effect of the pollen source on the maternal tissue might be mediated by the seeds, which serve as a source for plant hormones (Mizrahi et al., 2004). Swingle (1982) first raised this idea, when he tried to explain metaxenia in the date palm.

The effect of the pollen source on TSS (sugar content) and TA has been reported for several species from different families, including mandarins (Wallace and Lee, 1999), Vine Cacti (Mizrahi et al., 2004), apple (Bodoret al., 2008).

Other metaxenia effects that were found, e.g. on Antioxidant content , Total Flavonoid, Total Phenol, Vitamin C, Lycopene, β-carotene, pH and TSS/TA may also be explained by seed-controlled hormonal level. This again highlights that the role of plant hormones in metaxenia should be investigated (Mizrahi et al., 2004).

However, the effect of pollen source on fruit quality should be carefully examined since the pollen source might affect the taste qualities of the fruit. These results are similar to those reported for dates, cherimoya, grapes and mandarins, in which the source of pollen had an effect on maternal tissue characteristics (Denney, 1992; Wallace and Lee, 1999; Mizrahi et al., 2004).

Although the principal cause of metaxenia is not unambiguously understood, this phenomenon may be put to immediate use in horticultural practice (Mizrahi et al., 2004).

Conclusion

The heterosis, cytoplasmic and metaxenia effects occur in all qualitative characteristics of tomato fruit. This is the first report of metaxenia and cytoplasmic effects of qualitative traits in tomato. In this research, Heterosis, metaxenia and cytoplasmic effects are dependent on both the pollen source and female line. High amount of heterosis, based on mid parents was observed for lycopene and vitamin C. High amount of heterosis, based on the best parent was observed for lycopene. When antioxidant content was the desired end result, A line was the best female line, but when taste was desired, D line was the best female line. The best female line for antioxidant content, total flavonoid and total phenol was A line, while for TA was B line, for β-carotene and TSS% was C line and for vitamin C, lycopene, pH, and TSS/TA was D line. The best pollen source for total flavonoid, vitamin C and TA was A line, while for total phenol, lycopene, pH, TSS%, TSS/TA was B line, for β-carotene was C line and for antioxidant content was D line.

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