GENETIC STUDY OF ROCK MELON (CUCUMIS MELO L. VAR. CANTALUPENSIS):
FRUIT VARIABILITY IN SEGREGATING POPULATIONS AND AFTER SELFING PERFORMANCE

MARVELDANI, E. MAULANA, and R.B. NUGROHO*

Department of Food Crop Cultivation, Politeknik Negeri Lampung. Bandar Lampung, Lampung, Indonesia
*Corresponding author's email: rizkibagus23@gmail.com
Email addresses of co-authors: marveldani@polinela.ac.id, erie@polinela.ac.id

SUMMARY

Rock melon (Cucumis melo L. var. Cantalupensis) is a commercial-type melon with a wide distribution worldwide. Rock melons with larger fruit sizes are more in demand in restaurants, the food processing industry, and large families. In breeding for larger fruits, the increased genetic variability through introgression and recombination is the main factor; then, it can continue to purify before becoming a parental line. The presented study sought to recognize the variability of the open-pollinated population and their performance after selfing. The experiment ran from September 2021 to July 2022 at the Field Experimental Station, Politeknik Negeri Lampung, Indonesia. The experiment consisted of two sub-experiments carried out without experimental design. There was wide variability in both quantitative and qualitative variables in the open-pollinated populations, except fruit flesh color, weight, and sugar content. Population after selfing (S1 population) had more uniform qualitative traits. All the fruits sampled were medium elliptic with netted rinds followed by two-color variants (greenish yellow and green). Also, narrow variability occurred for the traits, fruit diameter, weight, and sugar content, followed by low heritability for the quantitative traits.

Keywords: Genetic variability, heritability, introgression and recombination, melon (Cucumis melo L.), open-pollinated population, quantitative and qualitative traits, selfed (S1 population)

Key findings: In F₂ segregating populations, a wide diversity of quantitative and qualitative traits proved that intercross ably maintained variability as per Handy-Weinberg equilibrium theory. Besides, the high heritability was evident for fruit shape and color traits after selfing, particularly for elongated and green color traits. They seemed like simple allelic traits.

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INTRODUCTION

Melon (Cucumis melo L., 2n = 2x = 24) is a herbaceous plant belonging to the Cucurbitaceae family. It is the most popular tropical fruit worldwide, known for its sweetness, fruit texture, and fragrance. The fruit also contains the best nutritional properties, such as, carbohydrates, protein, antioxidants (phenolic compounds, ascorbic acid, and carotenoids), flavonoids, alkaloids, vitamins (A, B6, and C), potassium, folate, niacin, and complex essential minerals like magnesium, sodium, and phosphorus (Huda et al., 2018; Manchali et al., 2021; Wibowo et al., 2021). In addition, melon seeds contain vitamin E and essential fatty acids.

The melon’s earlier domestication originated in Africa and Asia for lipid- and protein-rich seeds, followed by the consumers’ preference for its mesocarp after finding it sweet. Previous studies reported developing melons by selecting wild melons, which had thin and bitter flesh with tiny fruit, and the out-crossing nature of melons and the selection gradually turned melons into different horticultural usages (Manchali et al., 2021). The most common and recent use is for consuming tasteful and healthy fruit. Despite rapid developments in plant breeding technology, such as, molecular, metabolomics, omics, and genetic modification approaches (gene transfer and CRISPR genome-editing), its hybrid development through conventional hybridization remains the most common melon breeding program.

The hybrid cultivar developed by crossing two parental lines, although other methods use more parental lines, such as, hybrids from the double- and three-way. The parental lines should possess desirable traits that vary, with a higher value of general and specific combining abilities (Acquaah, 2012). Generally, the researcher needs a collection with a wide variation of the populations, then decides the desired traits for selection and purification until becoming the parental line. In melon, the hybrid-breeding process has a relatively high success rate, even if tedious. The reason is that melon has an extreme diversity of traits spread over the nine cultivar groups, i.e., C. melo var. Cantalupensis (cantaloupe/rock melon), C. melo var. Momordica (snap melon), C. melo var. Reticulatus (muskmelon), C. melo var. Inodorus (honeydew), C. melo var. Dudaim, C. melo var. Flexuosus (snake melon), C. melo var. Conomon, C. melo var. Chito, and C. melo var. Makuwa (oriental melon) (Suwarno et al., 2016; Huda et al., 2018). There were some ideotype-based options to breed a new parental line by using the melon diversity.

The extensive diversity exists not only among the cultivars but also within cultivars. The cultivar of C. melo, snap melon, has an extreme genetic variation for fruit traits, such as, shape, size, color, and sugar content (Rad et al., 2017; Mutheselvi et al., 2019; Kesh and Kaushik, 2021; Manchali et al., 2021). Like snap melon, rock melon also displays wide genetic variations (Yamaguchi et al., 1977; Jariani et al., 2021); however, there is no specific report, especially for its fruit traits’ variability.

Rock melon (C. melo var. cantalupensis), also called cantaloupe, is characterized an andromonoecious (containing male and hermaphrodite flowers in one plant) and a climacteric fruit. The cantaloupe melon is easily recognizable due to its slightly ribbed and net-like appearance. It has a relatively high sugar content, strong fragrance, short shelf-life, and juicy orange flesh (Suwarno et al., 2016; Fundo et al., 2018; Revanasidda and Belavadi, 2019; Manchali et al., 2021). Rock melon is also rich in vitamins (particularly A, B, and C), beneficial microelements (potassium and magnesium), and high levels of antioxidants. In addition, it has exhibited a hepato-protective effect activity against hypothyroidism and immune-modulating properties (Vella et al., 2019). Some genotypes of rock melon had higher stability and adaptability, including other promising traits to breed (Oliveira et al., 2019; Amarasinghe et al., 2021). Moreover, it adapts to extremely high temperatures (42 °C) and produces standard fruit quality (Amarasinghe et al., 2021). The genetic variation in segregating populations of rock melon is the key to breeding a desired cultivar for specific aims, such as, a larger fruit with high sugar.
content for restaurant necessities, food processing industry needs, and large families. The presented study sought to identify the fruit variability in the segregating populations and the population performance after selfing from the largest selected fruit.

MATERIALS AND METHODS

The segregating and S1 (selfed) populations developed from the F1 variety similar to Red Aroma, which has a green full-netted fruit rind, circular shape, and orange flesh, with high sugar content (approximately 12%), served as samples in this study, particularly the second and third planting seasons (Table 1). The study started from September 2021 to July 2022 at the Field Experimental Station of Politeknik Negeri Lampung, Indonesia (longitude and latitude at 5.3584° S and 105.2329° E, respectively). Seed soaking in warm water at 46 °C for 5 min commenced to speed up and increase the germination, controlled by a digital water thermometer. Seedling media consisted of a mixture of soil and manure with a ratio of 1:1, and black and white plastic mulches covered the soil. Four-week-old seedlings transplant continued into a 5 m² × 1 m² plot and spaced at 0.5 m² × 0.8 m². The 5 g per plant per two weeks of NPK fertilizer (N: P: K ratio was 16:16:16) application occurred during the plant vegetative stage, while SP-36 fertilizer and KCl (1 g per plant per week for five weeks) during the generative stage. An unblomed hermaphrodite flower (approximately 1 m from mulch) sustained a paper-based lid cover to encourage self-pollination. After the ovary had grown (2-3 days after flower treatment), covering with a fruit shield followed to protect the fruit from pests. Harvesting transpired when the fruit stalks turned brown.

The study consisted of two subexperiments. The first aimed to identify fruit variability in the segregating populations, while the second sought to calculate the genetic parameters in S1 populations. The first subexperiment comprised 720 plants of segregating populations, with 12% serving as representative samples. The second subexperiment used 300 plant-sized selfing populations, with 15% of the total population serving as the representative samples. Sample sizes and sampling design for quantitative measurement followed the method by Gomez and Gomez (1984) and Filho (2013), whereas qualitative data collection progressed on all plants in the populations.

These experiments’ implementation had no experimental design since both subexperiments used segregating populations. Qualitative traits observed in this study included mature fruit rind flesh color, fruit net, and shape in the longitudinal section. Qualitative trait identification relied on the CPVO (The Community Plant Variety Office) (2021) with the additional fruit shape identification formula based on the findings of Rashidi and Arabsalami (2016). This study observed the quantitative traits, such as, fruit length, diameter, longitudinal area, weight, and flesh sugar content. Fruit longitudinal area calculation used the ImageJ ver. 1.52a and measuring sugar content utilized a refractometer, Brix. In one fruit, four samples served to estimate the sugar content: one from the top end, one from the base end, and two from the middle of the fruit. These samples obtained averaging.

The collected data analysis included estimating the genetic variability and heritability. The genetic variability estimation continued by comparing the phenotypic variance ($\sigma^2_p$) with the standard deviation multiplied by 2(× SD); wide variability is when $\sigma^2_p > 2 \times SD$ (Nugroho and Anas, 2016). The estimation of broad sense heritability ($h^2_{bs}$) employed the formula of Merk and Foolad (2012):

$$h^2_{bs} = bop = CovOP/\sigma^2_p$$

Where:

bop = parent-offspring correlation
Cov OP = covariance parent-offspring
Table 1. Planting season scheme.

<table>
<thead>
<tr>
<th>Planting seasons</th>
<th>Populations</th>
<th>Treatments</th>
</tr>
</thead>
<tbody>
<tr>
<td>First</td>
<td>$F_2$ segregating / open-pollinated</td>
<td>Intercross</td>
</tr>
<tr>
<td></td>
<td>(seeds from commercial hybrid rock melon)</td>
<td></td>
</tr>
<tr>
<td>Second</td>
<td>Segregating / open-pollinated</td>
<td>Selfing</td>
</tr>
<tr>
<td>Third</td>
<td>Selfing 1 (S1)</td>
<td>Selfing</td>
</tr>
</tbody>
</table>

Table 2. Rock melon fruit color, net, shape variations, codes, and their proportion in the open-pollinated populations.

<table>
<thead>
<tr>
<th>Color</th>
<th>Net</th>
<th>Shape</th>
<th>Code</th>
<th>Proportion in population (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yellow</td>
<td>No net</td>
<td>Medium elliptic</td>
<td>A1</td>
<td>1.09</td>
</tr>
<tr>
<td>Yellow</td>
<td>Full net</td>
<td>Circular</td>
<td>A2</td>
<td>3.81</td>
</tr>
<tr>
<td>Yellow</td>
<td>Full net</td>
<td>Other</td>
<td>A3</td>
<td>2.72</td>
</tr>
<tr>
<td>Greenish-yellow</td>
<td>No net</td>
<td>Circular</td>
<td>B1</td>
<td>5.99</td>
</tr>
<tr>
<td>Greenish-yellow</td>
<td>No net</td>
<td>Other</td>
<td>B2</td>
<td>3.09</td>
</tr>
<tr>
<td>Greenish-yellow</td>
<td>Partial net</td>
<td>Circular</td>
<td>B3</td>
<td>2.18</td>
</tr>
<tr>
<td>Greenish-yellow</td>
<td>Partial net</td>
<td>Other</td>
<td>B4</td>
<td>3.45</td>
</tr>
<tr>
<td>Greenish-yellow</td>
<td>Full net</td>
<td>Circular</td>
<td>B5</td>
<td>8.17</td>
</tr>
<tr>
<td>Greenish-yellow</td>
<td>Full net</td>
<td>Medium elliptic</td>
<td>B6</td>
<td>1.63</td>
</tr>
<tr>
<td>Green</td>
<td>No net</td>
<td>Medium elliptic</td>
<td>C1</td>
<td>2.36</td>
</tr>
<tr>
<td>Green</td>
<td>No net</td>
<td>Other</td>
<td>C2</td>
<td>3.99</td>
</tr>
<tr>
<td>Green</td>
<td>Partial net</td>
<td>Circular</td>
<td>C3</td>
<td>3.27</td>
</tr>
<tr>
<td>Green</td>
<td>Partial net</td>
<td>Other</td>
<td>C4</td>
<td>0.54</td>
</tr>
<tr>
<td>Green</td>
<td>Full net</td>
<td>Circular</td>
<td>C5</td>
<td>22.14</td>
</tr>
<tr>
<td>Green</td>
<td>Full net</td>
<td>Medium elliptic</td>
<td>C6</td>
<td>2.72</td>
</tr>
<tr>
<td>Green</td>
<td>Full net</td>
<td>Other</td>
<td>C7</td>
<td>4.54</td>
</tr>
<tr>
<td>Dark green</td>
<td>No net</td>
<td>Circular</td>
<td>D1</td>
<td>4.36</td>
</tr>
<tr>
<td>Dark green</td>
<td>No net</td>
<td>Other</td>
<td>D2</td>
<td>1.45</td>
</tr>
<tr>
<td>Dark green</td>
<td>Partial net</td>
<td>Circular</td>
<td>D3</td>
<td>4.36</td>
</tr>
<tr>
<td>Dark green</td>
<td>Partial net</td>
<td>Other</td>
<td>D4</td>
<td>3.81</td>
</tr>
<tr>
<td>Dark green</td>
<td>Full net</td>
<td>Circular</td>
<td>D5</td>
<td>12.52</td>
</tr>
<tr>
<td>Dark green</td>
<td>Full net</td>
<td>Medium elliptic</td>
<td>D6</td>
<td>1.00</td>
</tr>
</tbody>
</table>

RESULTS

Fruit variability in segregating populations

The segregating population comprised genetically diverse progenies from a cross between two parental lines with distinct traits. A total of 551 fruits from 720 plants incurred observations in this study, then grouped (Table 2). The studies revealed that some plants showed abnormal growth, others did not bear fruits, and some even died. Some fruits sustained damage from pests and aborted during development, mostly occurring in the segregating populations. In the segregating rock melon populations, wide fruit variability was evident for qualitative and quantitative traits, except for fruit flesh color (Figure 1, Table 3), fruit weight, and sugar content (Table 3). It was also notable that some of the qualitative traits in the melon fruits had no description in the CPVO (2021), such as, fruits with greenish-yellow rind color, fruits with partial net, and fruits with ovate, oblate, and obovate shapes. The fruits determined with these shapes attained a grouping named "others" (Figure 1, Table 2). A devised simple code for grouping the fruit variation (Table 2) used the alphabet to denote the fruit rind color, while the sequential numbers indicated
both net and shape consecutively. The overall variability for the qualitative traits is available in Figure 1.

In the latest study, five quantitative fruit traits, i.e., fruit length, diameter, longitudinal area, weight, and sugar content, gained scrutiny. Wide genetic variability was apparent for the attributes, viz., fruit length, diameter, and area, and fruit weight and sugar content had narrow variability (Table 3). The distribution of the fruit type code appears in Figure 2. The wide variability observed in fruit length, diameter, and longitudinal area was in line with the qualitative traits of fruits (Figure 1). In simpler terms, the various types of fruits produced, such as, elongated and round fruits, will naturally affect the range of data in quantitative traits like fruit length and width. It was also noteworthy that small fruits contained less sugar content. Environmental factors and their interaction with the genetic makeup of the genotypes play a crucial role in fruit phenotype. High rainfall during the planting season was one of the main factors for nutrient loss that led to small fruit with less sugar content (Yao et al., 2021).

**Genetic parameters and performance of S1 populations**

Selfing from an open-pollinated plant like melon is the way to increase homozygosity for crossing purposes. The homozygous plant has the potential to become the succeeding

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**Figure 1.** Variability of fruit shapes, colors, and nets of rock melon in the segregating populations, (bar=10 cm).

**Table 3.** Variability of quantitative fruit traits in segregating populations.

<table>
<thead>
<tr>
<th>Traits</th>
<th>Mean</th>
<th>SD</th>
<th>2×SD</th>
<th>σ²</th>
<th>Variability</th>
<th>CV (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fruit length (cm)</td>
<td>17.94</td>
<td>2.66</td>
<td>5.31</td>
<td>7.06</td>
<td>Wide</td>
<td>14.81</td>
</tr>
<tr>
<td>Fruit diameter (cm)</td>
<td>15.84</td>
<td>2.26</td>
<td>4.51</td>
<td>5.09</td>
<td>Wide</td>
<td>14.24</td>
</tr>
<tr>
<td>Fruit longitudinal area (cm²)</td>
<td>236.61</td>
<td>64.11</td>
<td>128.23</td>
<td>4110.60</td>
<td>Wide</td>
<td>27.10</td>
</tr>
<tr>
<td>Fruit weight (kg)</td>
<td>1.35</td>
<td>0.38</td>
<td>0.76</td>
<td>0.14</td>
<td>Narrow</td>
<td>28.28</td>
</tr>
<tr>
<td>Sugar content (°Bx)</td>
<td>8.36</td>
<td>1.64</td>
<td>3.28</td>
<td>2.69</td>
<td>Narrow</td>
<td>19.60</td>
</tr>
</tbody>
</table>
parental line, resulting in a homogenous hybrid plant after crossing for commercial and research aims (Acquaah, 2012). Based on Figure 2, the heaviest fruits came from the segregating populations (from the C6 population) for planting in the next generation. The genetic study should proceed from the C6 typical fruit in the first selfing generation (S1 population). It was predictable to obtain the fruit size-controlling allele/gene after purifying this line for crossing purposes in the final breeding program.

Estimating genetic parameters is imperative to understanding the genotypes’ genetic basis, performance, and inheritance capability to determine the breeding strategy. The genetic parameters, such as, genetic variability and heritability estimation, appear in Table 4. A surprisingly narrow variability occurred for the fruit parameters, i.e., fruit diameter, weight, and sugar content, while the rest of the traits revealed broad variability.

In the S1 populations, all the fruits were medium elliptic with netted rind (Figure 3). For segregating populations, the fruits typically looked like B6 (medium elliptic, greenish-yellow, and netted rind) and C6 (medium elliptic, green, and netted rind) with a ratio of 28.5%:71.5% for B6:C6, respectively. In the S1 populations, C6 was longer and relatively larger (area and weight) than B6, having the same fruit shape. Moreover, C6 had relatively more sugar content than B6 (Figure 4).

**DISCUSSION**

For plant breeders, genetic variability plays a vital role in getting a desired and ideotype-based cultivar. *Cucumis melo* is known to have remarkable variations among and within cultivars (Suwarno et al., 2016; Manchali et al., 2021). Overall, and within cultivars, cultivar snap melon has extensive genetic and phenotypic variabilities (Rasool et al., 2016; Mutheselvi et al., 2019). Like snap melon, rock melon is likely to have wide phenotypic

Figure 2. Distribution of quantitative fruit traits with each fruit type code in segregating populations.
Figure 3. Variability of qualitative melon fruit traits in segregating populations, (bar=10 cm).

Figure 4. Distribution of quantitative fruit traits with each fruit type code in S1 populations.
variability, especially for fruit traits. Melon plants with desirable phenotypic traits attain selection and purification for several generations to increase the homozygosity and stability of the desired features. The preferred pure lines and ideotypes can become the parental lines for developing hybrid and open-pollinated cultivars, depending on their general and specific combining ability values (Reddy et al., 2007; Badami et al., 2020; Pratami et al., 2020).

Genetic variability can further sustain expansion through hybridization, mutation, protoplast fusion, and genetic transformation (Acquaah, 2012). Hybridization is the conventional approach commonly used in Southeast Asia. The second generation after hybridization, called F₂ (filial 2), has a maximum variability caused by segregating alleles/genes and heterozygosity in the populations. In F₂ populations, the breeder may encounter various seed conditions, including healthy or abnormal seed germination and non-germinating seeds caused by undesired and lethal gene accumulation (Acquaah, 2012). In developing countries, limited experimental areas often impede a comprehensive observation of the variability in the F₂ populations. By intercrossing the F₂s or the segregating populations planted in two planting seasons, trying to conserve, maintain, and bring out all the possible variabilities in the limited experimental area was prevalent. By selfing the segregating populations, the study aimed to improve the homozygosity of the desired traits in the next generation.

Fruit rind color attained effects from three metabolite groups, i.e., chlorophylls, carotenoids, and flavonoids. These metabolites cooperate to produce yellow, greenish-yellow, green, and dark green rind colors (Tadmor et al., 2010). When presenting a level above 10 μg g⁻¹ naringenin chalcone (4,2',4',6'-tetra hydroxyl chalcone), a yellow flavonoid pigment produces a yellow-colored rind. Feder et al. (2015) reported that the pigment has the CmkFB gene controlling it, a Kelch-domain containing F-box protein-coding gene located in chromosome 10. Naringenin chalcone’s production comes from the phenylpropanoid pathway by the regulation of phenylalanine ammonia-lyase (PAL), cinnamate 4-hydroxylase, and chalcone synthase (CHS, regulated by CmkFB). When naringenin chalcone (above 10 μg g⁻¹) interacts with a low level of carotenoid-chlorophyll (approximately 150–300 μg g⁻¹ for carotenoid and 25–50 μg g⁻¹ for chlorophyll), it results in a greenish-yellow rind. Green and dark green color emerges when the naringenin chalcone level is very low or undetectable, solely determined by carotenoid and chlorophyll levels. A combination of a medium level of carotenoid (approximately 300–600 μg g⁻¹) with a low level of chlorophyll (25–50 μg g⁻¹) results in green-colored rind. The dark green peel comes from a high level of both carotenoid and chlorophyll (above 600 μg g⁻¹ of carotenoid and 100 μg g⁻¹ of chlorophyll). The APRR2 transcription factor governs the level of expression of the carotenoid-chlorophyll.

Yellow was the rarest fruit rind color found in the melon populations, especially for no-net and medium elliptic fruits (Table 2). All the planted hybrids had green rinds, while the yellow skin started to appear in the segregating populations. The emergence of yellow rind fruits was likely due to the accumulation of naringenin chalcone and none of carotenoid-chlorophyll. The suspected crossover recombination during meiosis or nonsense mutation in the APRR2 transcription factor resulted in an insignificant carotenoid-chlorophyll level in the segregating populations.

Table 4. Genetic parameters of quantitative traits in the S1 populations.

<table>
<thead>
<tr>
<th>Traits</th>
<th>Mean</th>
<th>SD</th>
<th>2×SD</th>
<th>σ²p</th>
<th>Variability</th>
<th>CV (%)</th>
<th>h²w (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fruit length (cm)</td>
<td>22.44</td>
<td>3.24</td>
<td>6.49</td>
<td>10.53</td>
<td>Wide</td>
<td>14.46</td>
<td>0</td>
</tr>
<tr>
<td>Fruit diameter (cm)</td>
<td>16.70</td>
<td>1.30</td>
<td>2.60</td>
<td>1.69</td>
<td>Narrow</td>
<td>7.78</td>
<td>0</td>
</tr>
<tr>
<td>Fruit longitudinal area (cm²)</td>
<td>305.74</td>
<td>51.43</td>
<td>102.86</td>
<td>2644.91</td>
<td>Wide</td>
<td>16.83</td>
<td>0</td>
</tr>
<tr>
<td>Fruit weight (kg)</td>
<td>1.72</td>
<td>0.27</td>
<td>0.54</td>
<td>0.07</td>
<td>Narrow</td>
<td>15.79</td>
<td>0</td>
</tr>
<tr>
<td>Sugar content (°Bx)</td>
<td>8.37</td>
<td>1.60</td>
<td>3.21</td>
<td>2.58</td>
<td>Narrow</td>
<td>19.16</td>
<td>5.49</td>
</tr>
</tbody>
</table>

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populations, thus producing yellow rind fruits. According to Tadmor et al. (2010), naringenin chalcone also emerged in the greenish-yellow rind. Around 30% of the plants in the population exhibited the accumulation of naringenin chalcone, represented by yellow and greenish-yellow colored peels (Table 2). The largest and second-largest rind color proportion was green and dark green, respectively (Table 2). The share for fruit rind color inheritance was 8:5:5:2 (green: dark green: greenish-yellow: yellow). Only a few fruits producing naringenin chalcone in segregating populations surfaced, which likely resulted in the crossing between green × greenish-yellow lines. Further chemical tests and molecular approaches were necessary to confirm this hypothesis.

In segregating populations, all the fruits had orange flesh. Orange flesh has one specific gene controlling it called CmOr; it is responsible for the accumulation of β-carotene (Tzuri et al., 2015; Oren et al., 2019). A G/A single nucleotide polymorphism (SNP) at the position of the 323rd nucleotide of this gene associates with flesh color, where G is responsible for a green or white flesh color, and A is responsible for the orange color. Sedera et al. (2016) and Selim (2020) reported that the gene action of orange flesh was evident with complete dominance of high heritability. All progeny will have orange flesh if both parental genotypes are dominant CmOr\textsuperscript{orange}.

The segregating populations had fruits with partial net discovered (Figure 1), and it aligns with the findings of Oren et al. (2019). Fruits (22.33%) in the population showed without rind net, 17.61% with partial, and 60.06% with full net rind (Table 2). It has a 1: 1: 3 (for no-net: partial net: full net) proportion, respectively. This proportion followed the Mendelian inheritance if the no-net allele was recessive. We assumed that one parental line had a smooth rind and the other was a full-net rind. A portion of the plants from this segregating population produced fruits lacking net rinds, and it might signify the roles of quantitative trait loci (QTLs) governing smooth skins. At least six rinds net-associated QTLs have attained mapping in several linkage groups (LGs): LG I, LG II (one major QTL ntc2.1 and three QTLs for net density, i.e., ntd2.1, ntd2.2, and ntd2.3), LG III, LG XI (minor QTL ntc11.1), and LG XII (minor QTL ntc0812.1). Past studies also reported the involvement of several other interacting QTLs for this trait (Beja et al., 2010; Puthmee et al., 2013; Vegas et al., 2013; Oren et al., 2020).

Net or reticulation is a network of suberized periderm tissue responding to the cracking of the fruit surface during enlargement. While the melon's ancestral phenotype is a smooth rind, an independent dominant mutation caused a rise to netted fruit, which later became widely domesticated. The wide variability of net rind patterns in the segregating population has the control of multiple genes, as well as mechanical, physiological, and metabolic factors (Oren et al., 2020). Generally, netted melon is also climacteric even though fruit net has an association with peroxidase (POD) activity and climacteric has influences from ethylene levels. It resulted in a linkage between genes controlling fruit net and two climacteric ethylene in LG III. Huda et al. (2018) also reported that melons with netted rinds had higher ethylene levels than smooth ones. In addition, high ethylene level leads to orange flesh. Contrary to those studies, we found net and shiny rind fruits with orange flesh in our population. These findings indicated the complex relationship between the melon’s net, ethylene, and flesh color.

In the presented study, the proportion of circular: medium elliptic: “others” was visible for fruit shape in the segregating population with a ratio of 7:1:2. Even in this population, the variability of fruit shape was wide; most of the fruits were shaped circular, much like the F\textsubscript{1} population. Medium elliptic and “other” forms were of equal distribution in the population, likely due to the segregation of some minor genes. Hybridization between two circular parental lines could still produce this proportion. Melon fruit shape studies have been numerous, with some QTLs identified and mapped underlying such diversity. Several alleles are known to govern specific fruit shapes, such as, the CALC allele for elongated or medium elliptic fruit, CALC8-1 allele for
round or circular fruit, PS allele for oval, and CmFS18/CmOFP13 for ovate fruit (Martinez et al., 2022). The CmTRMS, CmSUN12, CmCRC, and other genes correlate with fruit size and shape (Diaz et al., 2014; Ma et al., 2022; Martinez et al., 2022). Fruit shapes depended on the expression level of those genes and correlated with ovary shape (Ramamurthy and Waters, 2015). The role of multiallelic contributed to the maximum variability in the F2 segregating populations (Acquaah, 2012).

Recent studies have proven that some qualitative traits have bi or multiallelic control, rarely by monoallelic. The qualitative traits incurred a common quantification approach, as Oren et al. (2020) reported. In the study, they set nine levels of rind density variability from their collection. Specific gene roles sometimes release an intense effect on particular traits, while others are generally complicated. Some qualitative traits in melon, like color, netting, and shape, showed a complex expression pattern. Nevertheless, qualitative traits are faster to breed and develop than quantitative traits as controlled by multiple alleles, environmental, and genetic by environmental (G × E) factors (Acquaah, 2012).

The C6 population produced the bulkiest fruits (length, area, and weight), while the B2 population had relatively the tiniest fruits compared with the other populations. The C5 and D3 populations had the highest sugar content, while B2 was in contrast. The B2 population displayed poor genetic performance and was far from the market criteria. Fruit shape and size (length, diameter, and weight) were integral, having control from additive and epistatic gene interaction (Huda et al., 2018; Lian et al., 2021). Diaz et al. (2014) mapped some QTLs for fruit size. Fruit length was distinct at LG II, IIIb, VIa, and VIII, and fruit diameter was apparent at LG IIIa and XII. The QTL for sugar content has central regulation from the Brix9-2-5, with the other QTLs found at LG VIII, IX, and X (Diaz et al., 2014; Pereira et al., 2018). Even though these traits had the control of multiple nuclear expressions, some candidate genes for early identification had been reported, such as, MELO3C025758 for fruit size and Brix9-2-5 for sugar content (Lian et al., 2021).

The S1 population selected from C6 with the heaviest fruits produced medium elliptic shapes. The shape attained most inheritance from the previous generation. Fruit shape revealed correlatedness with the ovary shape, as controlled by the dominance effect (Ramamurthy and Waters, 2015). In S1 populations, no yellow and dark green rind indicated that APRR2 was always in expression without over-expression. Yet, a greenish-yellow color emerged in this population. We assumed that a slight expression level of CmkFb in the segregating population of C6 resulted in a poor yellow expression that was most probably imperceptible during the visual observation; however, it accumulated in some plants in the S1 population. The fruits’ flesh color was orange, indicating no interference to gene expression and no mutation of CmOForange happened; thus, orange flesh still dominated the population. Aside from flesh color, the net pattern was also a complete inheritance from the previous generation, probably for the same reason in addition to the high heritability of this trait (Oren et al., 2020).

In the S1 population, the homozygous-like state for some qualitative traits and the narrow variability for some quantitative traits also had previous reports (Amorim et al., 2016). Strong major QTL expression could be the reason, followed by no mutation and crossing over. Strongly inheriting the CALC and PI124112 alleles resulted in the elongated fruit with narrow variability of fruit diameter. The limited variability of fruit weight and sugar content as controlled by multi-allele expression was probably due to the heavy rain during the planting season. Planting in a greenhouse could further result in a more ideal variability, where containing the environmental effect could be better. Aside from that, no high broad sense heritability was notable among the traits, confirming that continual line purification should proceed in the next selfing population.

CONCLUSIONS

Segregated population analysis can estimate hybrid parental line traits. The study estimated that if the double cross approach were in
execution, the hybrid would result from the combination of green × greenish-yellow fruit color, smooth × full net rind, and circular × circular fruit shape, or vice versa. Intercross can maintain potential genetic variability from F2 seeds. The highest variability values were evident in almost all qualitative and quantitative traits. The selected fruits (the largest found from the C6 population) can strongly inherit major qualitative features. Comprehensive analysis using molecular markers will be a vastly engaging attempt to answer various questions in this study.

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REFERENCES


Tzuri G, Zhou X, Chayut N, Yuan H (2015). A “golden” SNP in CmOr governs the fruit...
flesh colour of melon (*Cucumis melo*). The Plant J. 82: 267-279.


