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## Fruit characteristics of F1 NIL *Sletr1-2* vary with parental background

**Abstract.** The *Sletr1-2* allele, which reduces ethylene sensitivity in tomatoes, offers a promising approach to extending fruit shelf life by slowing ripening-related metabolism. However, the trait's success depends on compatibility with the recipient cultivar's genetic background. This study evaluated fruit characteristics of F1 near-isogenic line (NIL) hybrids carrying *Sletr1-2* in the commercial backgrounds 'Costoluto', 'Oxheart', and 'Sart Roloise'. The field trial was carried out between May and August 2024 at the Experimental Station of Universitas Padjadjaran, Sumedang, Indonesia, following a randomized complete block design with eight biological replicates for each treatment. The results revealed that fruit characteristics of F1 NIL *Sletr1-2* hybrids depend on parental background: NIL *Sletr1-2* x 'Oxheart' produced small fruit size, thin pericarp, and poor visual quality, while NIL *Sletr1-2* x 'Costoluto' and NIL *Sletr1-2* x 'Sart Roloise' showed more commercial performance, with bigger fruit size. These findings highlight the critical role of parental background in optimizing *Sletr1-2*-linked traits and inform breeding strategies that balance postharvest resilience with commercial quality.

**Keywords:** Ethylene · Fruit · Postharvest · Tomato

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## Introduction

The increasing frequency of extreme weather events associated with global climate change has posed significant challenges to agricultural productivity, particularly in horticultural crops such as tomatoes (*Solanum lycopersicum*) (Singh et al., 2022). As climacteric fruits with a high water content (~94% of their total weight), tomatoes are highly susceptible to postharvest deterioration due to their elevated respiration rates and accelerated ripening, especially under heat-stress conditions. (Alenazi et al., 2020; Mubarok, Okabe, et al., 2015). In response to these challenges, recent research has focused on breeding strategies to develop tomato cultivars with extended fruit shelf life.

Hybrid tomato breeding has shown considerable promise in improving both yield potential and fruit quality. However, a critical limitation remains in integrating desirable traits, particularly abiotic and biotic stress tolerance, along with postharvest longevity, without compromising agronomic performance. (Patanè et al., 2021). Advances in tomato genetics have identified the *SlETR1* gene, which encodes an ethylene receptor, as a key regulator of ethylene-mediated responses. (Saadah et al., 2023). A specific allele, *Sletr1-2*, has been associated with reduced ethylene sensitivity, delayed ripening, and extended shelf life while maintaining essential agronomic traits. (Mubarok et al., 2023).

The *Sletr1-2* mutation, located within the second transmembrane domain of the ethylene receptor, disrupts ethylene perception, resulting in firmer fruit and approximately 4–5 days of prolonged shelf life compared to wild-type lines (Saito et al., 2011). Despite its potential for postharvest improvement, mutant lines carrying *Sletr1-2* often exhibit agronomic drawbacks, including smaller fruit size and higher acidity, which limit their commercial appeal (Mubarok et al., 2016).

To overcome these limitations, the *Sletr1-2* allele can be introgressed into elite cultivars through the development of Near-Isogenic Lines (NILs). This approach employs repeated backcrossing between a donor parent carrying the target gene and a recipient parent with superior agronomic traits, resulting in NILs that retain the genetic background of the elite cultivar while incorporating the beneficial mutation (Di Giacomo et al., 2020). Hybrid F1 NILs carrying *Sletr1-2* are thus expected to combine stress

tolerance and shelf-life extension with desirable fruit quality attributes from the elite parental lines.

Nonetheless, the successful application of this breeding strategy depends on the genetic compatibility between *Sletr1-2* mutants and the commercial parental backgrounds. Incompatibilities can lead to undesirable physiological interactions, such as impaired heat tolerance or reduced fruit quality traits, namely size, flavour, and nutritional content. (Nguyen et al., 2019). These potential trade-offs underscore the necessity for a comprehensive evaluation of fruit quality parameters and secondary metabolite profiles in F1 NIL *Sletr1-2* hybrids derived from diverse genetic backgrounds. Therefore, the present study aims to evaluate and compare the fruit characteristics of F1 NIL *Sletr1-2* hybrids derived from different parental lines.

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## Materials and Methods

**Plant Materials and Cultivation.** Near-isogenic lines (NILs) carrying the *Sletr1-2* allele were crossed with three homozygous commercial tomato cultivars, 'Costoluto', 'Oxheart', and 'Sart Roloise', to produce F1 hybrid progenies. The field experiment was conducted from May to August 2024 at the Universitas Padjadjaran Experimental Station, Sumedang, Indonesia, using a randomized complete block design with eight biological replicates per treatment.

Seeds were germinated in seedling trays containing a 1:1 (v/v) mixture of cocopeat and rice husk charcoal. At the 5–6 true leaf stage, seedlings were transplanted into field plots and fertilized with NPK 15:15:15 at a total rate of 600 kg ha<sup>-1</sup>, applied in three split doses: 50% at transplanting, 25% at three weeks after planting (WAP), and the remaining 25% at six WAP. All plants were maintained under uniform agronomic practices throughout the growing season.

**Pericarp Thickness.** To evaluate internal fruit morphology, three fruits per plant at the breaker plus six days (Br+6) stage were cut transversely, and the number of locules was recorded and averaged. Pericarp thickness was measured at the equatorial region of each fruit using a digital caliper and expressed in millimeters (mm). The fruit-set percentage was calculated as the ratio of the total number of fruits to the total number of flowers per plant and served as an indicator of reproductive success.

**Titrateable Acidity (TA).** Titrateable acidity was determined following the procedure by Mauro et al. (2020). Five g of the homogenized tomato sample were diluted with 50 mL of distilled water, and the solution was titrated with 0.1 N NaOH until the pH reached 8.1. TA (%) was calculated using the formula:

$$TA = \frac{mL\ NaOH \times N\ (NaOH) \times 0.0064 \times 100}{g\ sampel}$$

**Total Soluble Solids (TSS).** TSS content was measured using a handheld refractometer (Euromex, Arnhem, Netherlands). A 5 g fruit sample was blended, centrifuged at 1,000 rpm, and 1000 µL of the supernatant was placed onto the refractometer lens. Measurements were performed in triplicate for each sample.

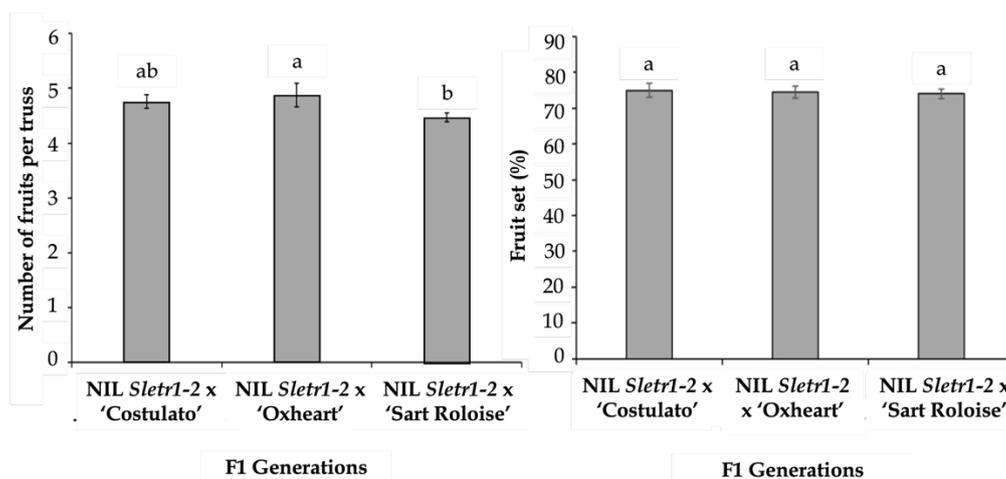
**Statistical Analysis.** All quantitative data were subjected to one-way analysis of variance (ANOVA), and treatment means were compared using Duncan’s Multiple Range Test (DMRT) at a 5% significance level. Statistical analyses were conducted using SPSS software version 25.0 (IBM Corp., Armonk, NY, USA).

## Results and Discussion

**Number of Fruits per Truss and Fruit Set.** The F1 NIL *Sletr1-2* hybrids showed variation in fruit number per truss, with statistically significant

differences among parental backgrounds. The average number ranged between 4.47 and 4.87 fruits per truss (Figure 1). The hybrid derived from ‘Oxheart’ produced the highest fruit number (4.87), although not significantly different from that of the ‘Costoluto’ (4.75). The cross with ‘Sart Roloise’ resulted in the lowest fruit number (4.47). These differences may reflect variations in inflorescence architecture and flower fertility influenced by parental genetic contribution. The number of fruits per truss influenced the plant yield. Mubarok et al. (2023) reported that fruit weight was affected by the number of fruits on the plant.

Fruit set is defined as the ratio of the number to the number of flowers. The reduction in fruit set had a consequential impact on plant yield (Mubarok et al., 2023). The statistical data analysis showed that the fruit ratio among the F1 NIL *Sletr1-2* was comparable. The values ranged from 74% (‘Sart Roloise’) to 75% (‘Costoluto’) (Figure 1). The consistent fruit set among all parental backgrounds suggests that the introgression of *Sletr1-2* did not adversely affect pollination efficiency or fertilization success under field conditions, and that all hybrid lines had comparable reproductive performance. Our previous study reported that environmental factors, such as temperature, significantly affect fruit formation (Mubarok et al., 2023). Ezura et al. (2019) reported that heat stress increased pollen sterility, leading to pollen development failure and pollination failure.



**Figure 1.** Number of fruits per truss and fruit set in various F1 NIL *Sletr1-2* tomato plants from three parental backgrounds (‘Costoluto’, ‘Oxheart’, and ‘Sart Roloise’). The average values followed by the same letter are not significantly different based on Duncan’s Multiple Range Test at the 5% level.

### Locule Number and Pericarp Thickness.

Locule number, which reflects the internal complexity and potential seed-bearing capacity of the fruit, varied significantly between hybrids (Figure 2). The F1 NIL *Sletr1-2* from 'Sart Roloise' parent produced the most locules, averaging 4.98 per fruit, indicating a more segmented internal fruit architecture. This was followed by the NIL *Sletr1-2* x 'Costoluto' fruit at 4.58, and finally, the NIL *Sletr1-2* x 'Oxheart' resulted in the lowest locule number at 4.28. Although all fruits fell within the typical multilocular category, the degree of locule formation differed in a way that highlights the influence of each parent line's inherent fruit morphology on this trait.

Pericarp thickness is a structural trait associated with fruit firmness. Among the hybrids, significant differences were observed (Figure 2). The F1 NIL *Sletr1-2* from 'Costoluto' parental background displayed the thickest pericarp at 3.98 mm, providing a potential advantage for mechanical strength and water retention. The F1 NIL *Sletr1-2* from 'Sart Roloise' parental background followed closely with a pericarp thickness of 3.73 mm, while the F1 NIL *Sletr1-2* from 'Oxheart' parental background had the thinnest pericarp, averaging only 3.11 mm. These results indicate that pericarp thickness is significantly influenced by the genetic contribution of the commercial parent and that selecting the parental line is crucial for determining this attribute.

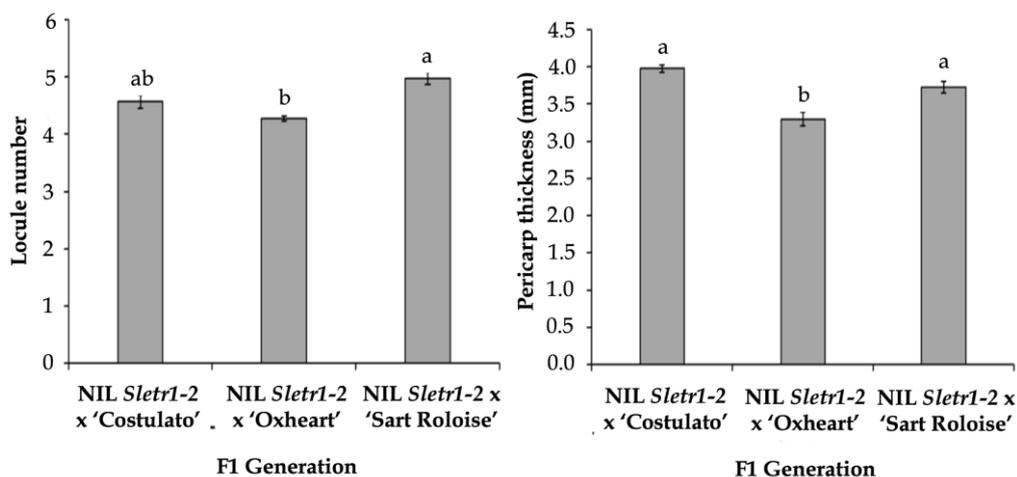


Figure 2. Locule number and pericarp thickness in various F1 NIL *Sletr1-2* tomato plants from three parental backgrounds ('Costoluto', 'Oxheart', and 'Sart Roloise'). The average values followed by the same letter are not significantly different based on Duncan's Multiple Range Test at the 5% level.

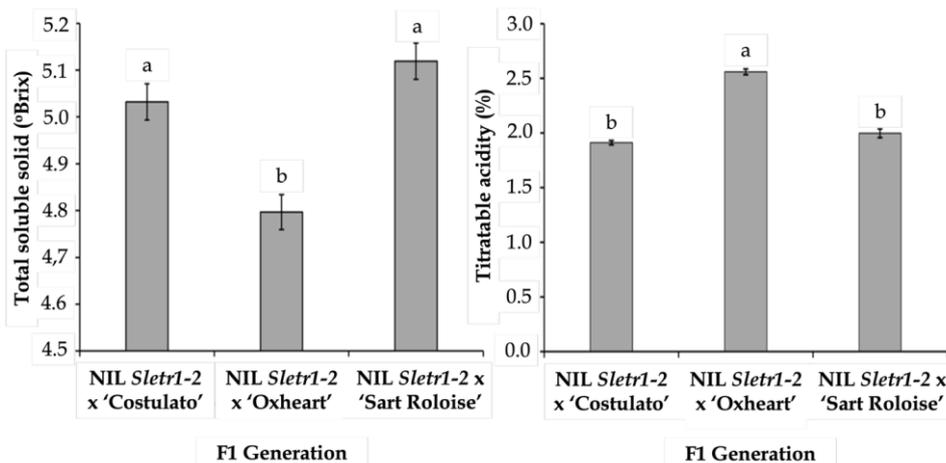


Figure 3. TSS and TA in various F1 NIL *Sletr1-2* tomato plants from three parental backgrounds ('Costoluto', 'Oxheart', and 'Sart Roloise'). The average values followed by the same letter are not significantly different based on Duncan's Multiple Range Test at the 5% level.

**Total Soluble Solids (TSS) and Titratable Acidity (TA).** TSS values, indicative of sugar content and flavor, ranged from 4.80 to 5.12 °Brix across hybrids (Figure 3). The NIL *Sletr1-2* × ‘Sart Roloise’ had the highest TSS (5.12 °Brix), which was significantly greater than the NIL *Sletr1-2* × ‘Oxheart’ hybrid (4.80 °Brix), but not different from the NIL *Sletr1-2* × ‘Costoluto’ (5.02 °Brix). These results highlight that the parental background influences fruit sweetness and that the *Sletr1-2* introgression does not suppress soluble sugar accumulation.

Titrate acidity is an indicator of sourness. Statistical analysis showed a significant difference among the F1 NIL *Sletr1-2* hybrid lines (Figure 3). The NIL *Sletr1-2* × ‘Oxheart’ had the highest TA value at 2.56%, indicating a more acidic flavor profile. In comparison, the NIL *Sletr1-2* × ‘Costoluto’ and × ‘Sart Roloise’ recorded significantly lower acidity levels at 1.91% and 2.00%, respectively. These differences reflect how each parent's genetic contribution affects organic acid biosynthesis and accumulation, thereby influencing overall flavor perception. The fruit flavor is influenced by the combination of sweet and acidic. TSS and TA are mostly used to estimate sugar and organic acids, respectively (Mubarok et al., 2015). In our study, we reported that the TSS and TA of F1 NIL *Sletr1-2* hybrid lines are dependent on the parental background. The parental line that crosses with NIL *Sletr1-2* will affect the fruit taste quality and fruit characteristics. This finding was similar to the previous study by Mubarok et al. (2015), the effect of *Sletr1-2* mutation was dependent on the genetic background of the pure-line cultivar parent.

Introducing the *Sletr1-2* allele into commercial tomato lines offers a practical strategy to improve shelf life by reducing the plant's sensitivity to ethylene, the hormone responsible for triggering ripening. By slowing ethylene-mediated metabolic activity, this mutation extends the fruit's postharvest life (Rahmat et al., 2021). However, the success of this approach depends not only on the mutation itself but also on its interaction with the genetic background of the recipient cultivar (Mubarok, et al., 2015). In this study, F1 NIL *Sletr1-2* hybrids were developed using three different parental lines: Oxheart, Sart Roloise, and Costoluto. The results showed apparent differences in fruit quality, postharvest potential, and metabolite

content depending on the chosen parental background (Saadah et al., 2023).

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## Conclusion

This study demonstrates that parental background strongly influences the expression of fruit characteristics in F1 NIL *Sletr1-2*. The ‘Oxheart’ background tended to amplify less desirable attributes, including lower pericarp thickness, lower TSS, higher acidity (TA), and smaller fruit size, which collectively reduce firmness, sweetness perception, and postharvest durability. These features make ‘Oxheart’ a less suitable candidate for commercial improvement when combined with the *Sletr1-2* allele. In contrast, ‘Costoluto’ and ‘Sart Roloise’ contributed to improved fruit size. Integrating ethylene-insensitive traits into elite tomato backgrounds requires strategic parental selection to balance storability, fruit size, flavour, and consumer-preferred qualities. These results demonstrate that integrating ethylene-insensitive traits such as *Sletr1-2* into elite tomato backgrounds requires careful parental selection to balance extended storability with desirable consumer-oriented attributes, including sweetness, firmness, and optimal fruit size.

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