Applied Fruit Science (2025)67:177
https://doi.org/10.1007/s10341-025-01375-x

ORIGINAL ARTICLE

Mitigating Fruit Cracking of Sweet Cherries Through *Ascophyllum nodosum*, Glycine Betaine, Calcium Chloride, and Silicon Treatments

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Received: 20 October 2024 / Accepted: 20 April 2025
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Abstract
Preharvest foliar treatments have become a widely used practice in various horticultural crops to improve their adaptation to rapidly changing climatic conditions. In light of these unstable climatic patterns, sweet cherry cracking has emerged as a critical problem that significantly affects the market value of fruit and yield. The objective of this study was to evaluate the effects of *Ascophyllum nodosum* (AN), glycine betaine (GB), calcium chloride (Ca), and silicon (Si) treatments on the cracking index (%) and overall quality of the sweet cherry cultivar 'Royal Tiago.' The Si, Ca, and GB treatments resulted in a significant reduction in the cracking index by 71.00%, 35.38%, and 26.00%, respectively, compared to the control. In contrast, AN treatment increased the cracking index by 26.12%. All preharvest treatments significantly increased the firmness of sweet cherry fruit. Notably, the Pearson's correlation coefficient showed a negative correlation between cracking index and antioxidant activity in 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay (r = –0.69) as well as between fruit firmness (r = –0.61), total phenolic content (r = –0.75), and acidity (r = –0.57). Ca and Si treatments significantly increased malic acid content and improved the organic acid profile of the fruit. In addition, AN, GB, and Ca foliar fertilizers significantly contributed to increase sugar, organic acids, total anthocyanin content, fruit weight, fruit firmness, and DPPH. The results obtained provide valuable insights into the control of fruit cracking and also appear to be a low-cost and environmentally safe means of improving the quality and marketability of sweet cherry fruit as well as facilitating harvest and postharvest management.

Keywords Biochemical constituents · Foliar treatment · Fruit firmness · Fruit quality · Biostimulants

Introduction

Cultivation of sweet cherries (*Prunus avium* L.) increased steadily worldwide during the past decade and in 2022, total world production reached 2.7 million tons. Turkey is currently one of the world's leading producers of sweet cherries, with a cultivated area of 80,482 ha and a total production of 656,041 tons in 2022 (FAO 2020). Sweet cherries have considerable economic value due to their desirable commercial characteristics, high nutritional content, and associated health benefits. The growing demand for sweet cherries has gradually highlighted the importance of storage potential and fruit quality. Recent climate projections indicate a decrease in water availability, an increase in air temperatures, and the occurrence of extreme events such as excessive rainfall close to the harvest period, which could lead to an increased incidence of fruit cracking in sweet cherries (Correia et al. 2020). This, in turn, results in significant economic losses due to a significant reduction in the commercial value of sweet cherry fruit.

Two primary causes of rain cracking in sweet cherries have been identified. One is excessive water uptake by the fruit skin, either directly or through the tree's vascular system, which leads to cracking (Correia et al. 2018), and the other is water adhesion to the fruit surface, which causes microcracks to expand into visible cracks (Winkler et al. 2020).

The intensity of cracking is a primary standard for grading fruit quality. The absence of cracking (sound fruit) corresponds to quality class "extra," while fruit with slight lateral cracking is assigned to class I. In this perspective, strategies that reduce the incidence and intensity of cracking will significantly improve fruit quality, storage potential, and shelf life (Karantzi et al. 2021).

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Published online: 13 June 2025

Development of existing methods to mitigate sweet cherry fruit cracking, such as breeding crack-resistant cultivars, has faced significant challenges, particularly in terms of identifying and modifying genes associated with crack resistance while maintaining fruit quality (Quero-García et al. 2021). Physical interventions, such as the use of rain covers, have shown limited effectiveness in reducing rain-induced cracking in sweet cherries (Knoche and Winkler 2017). Given the commercial importance of certain qualitative traits, sweet cherry growers, as well as packers and exporters, are eager to adopt strategies that improve external quality, including larger fruit size, dark and uniform skin color, absence of surface defects due to cracking, and firm fruit flesh.

Various compounds have been used in orchards to improve cherry quality and yield while reducing chewing through targeted plant nutrition strategies (Tsantili et al. 2007). Calcium plays a crucial role in cell wall formation and is a key determinant of the mechanical properties of plant tissues; it is also the most extensively studied nutritional factor in relation to fruit cracking (Santos et al. 2023). Glycine betaine (GB) is a quaternary ammonium compound that accumulates in a variety of organisms that thrive in arid or high-salt environments. It acts as a non-toxic, cell-compatible solute that reduces the osmotic potential within cells under stress-induced hypertonic conditions (Mansour 1999). In addition, GB is more effective than other compatible solutes in stabilizing membrane integrity, enzymes, and functional units such as the oxygen-evolving PSII complex and rubisco (Allakhverdiev et al. 2003). Based on the literature, GB increased the quality of sweet cherries and delayed fruit ripening (Gonçalves et al. 2020). Currently, detailed information on the effect of GB on sweet cherry cracking is lacking. Some studies have investigated the potential effects of biostimulants, such as seaweed-based products (Correia et al. 2015), on fruit quality. Biostimulants can be applied in the context of sustainable agriculture, as they are considered inputs with minimal environmental impact (Rasouli et al. 2022; Mannan et al. 2023; Zenobi et al. 2024). Seaweed-based biostimulants, such as *Ascophyllum nodosum* (AN), are known for their efficacy in horticulture, providing growth-promoting benefits and improving crop resilience to abiotic stresses such as salinity, temperature extremes, nutrient deficiencies, and drought (Asadi et al. 2022). The chemical composition of AN extract includes complex polysaccharides, fatty acids, vitamins, phytohormones, and essential minerals (Battacharyya et al. 2015). Several studies have reported that biostimulants increase plant productivity by increasing mineral assimilation, increasing the photosynthetic rate, reducing transpiration, and decreasing the incidence of fruit cracking (Correia et al. 2020; Gonçalves et al. 2020; Santos et al. 2023). On the other hand, the role of silicon in agriculture has become a focus of research due to its many benefits for plants, although it is not considered essential for most vascular plants (Spalevic et al. 2022; Shahzad et al. 2022; Irfan et al. 2023; Kumar et al. 2023). The application of silicon is increasingly seen as a promising, sustainable, and environmentally friendly approach to managing a wide range of plant diseases while enhancing plant growth and yield (Etesami and Jeong 2020). The beneficial effects of silicon on yield and quality have been documented in various horticultural crops, including apple (Karagiannis et al. 2021), avocado (Dann and Le 2017), and tomato (Xue et al. 2012). To the best of our knowledge, there is no information on the effect of this element on the cracking index and quality of sweet cherries.

'Royal Tiago' is an early-ripening sweet cherry cultivar harvested in May in the Çukurova region of the eastern Mediterranean region of Turkey, with a chilling requirement of 500 h. In this region, prolonged and heavy rains during this critical period often led to fruit cracking (Kuden et al. 2022). Therefore, this study is particularly important to improve the cultivation of sweet cherries under such conditions. The aim of this study is to evaluate the effects of preharvest treatments including calcium chloride ($CaCl_2$), glycine betaine (GB), seaweed extract from *Ascophyllum nodosum* (AN), and silicon (Si) on the cracking index (%) and fruit quality of 'Royal Tiago' sweet cherries produced in Adana, Turkey.

Materials and Methods

Plant Material

Six-year-old self-fertile (S1–S4) sweet cherry (*Prunus avium* L.) 'Royal Tioga' trees grafted on MaxMa 14 (*Prunus mahaleb* × *P. avium* L.) rootstock were established in an orchard at Çukurova University, Adana (37°01′41″N latitude, 35°21′59″E longitude, 50 m altitude). The trees were cultivated with a distance of 4 × 2.5 m between and within the rows, and the trees were pruned with the Kym Green Bush (KGB) system. The orchard was subjected to regular fertilization and irrigation.

Experimental Design

The study used a randomized block design with three blocks. For each block, five trees were selected for each treatment. These treatments were GreenStim, a proprietary liquid fertilizer combined with glycine betaine (GB; Miller Chemical & Fertilizer, USA); Zumsil Plus (Si; an advanced silicon technology formed by low-molecular-weight molecules; Zoberbac Agrocompany S.L., Spain); *Ascophyllum nodosum* (AN; 0.05%); $CaCI_2$ (Ca; 0.5%);

and control (CO; water). The dosages for each treatment were as follows: for every 100 l of water, mix 400 g of GreenStim, 400 ml of Zumsil Plus, 100 g of *Ascophyllum nodosum*, and 500 ml of calcium chloride ($CaCl_2$). Only water was used for the control.

Two buffer trees were placed between each sprayed tree to avoid any potential interference during the treatment process. Spraying was conducted 3 weeks before harvest, when the fruit had reached a yellow-straw color. Treatments were applied in the morning under dry and windless conditions using a low-pressure backpack sprayer. During the commercial harvest on May 3, 2024, 100 fruits were hand-harvested from each experimental tree to evaluate the effectiveness of the treatments in reducing fruit cracking and for subsequent pomological and quality analysis. The harvested fruits were then transported in a refrigerated box to the Post-Harvest Physiology Laboratory at the Department of Horticulture, Faculty of Agriculture, Çukurova University, where the analyses were performed.

Pomological Measurements

The average fruit weight (g) and seed weight (g) were determined using a digital balance with an accuracy of 0.01 g. The average fruit width (mm), fruit length (mm), fruit height (mm), and stem length (mm) were measured using a digital caliper with an accuracy of 0.01 mm. A total of 45 fruits (3 replicates of 15 fruits each) were used for measurements. Fruit firmness was measured at the equatorial region of the fruit using a Lloyd LF Plus texturing device (Ametek, UK) with a 3-mm diameter probe, applying a force of 50 N at a constant speed of 100 mm/min. To determine the acid and Brix content of fresh fruit juice, 0.2 ml of fruit juice was diluted with 10 ml of pure water and added to the reservoir of a digital Brix-acid meter (Atago PAL-BX, Japan), where measurements were taken directly by the instrument.

Color Measurements

Fruit peel color was measured in terms of CIE *L, a, b* using a colorimeter (CR-300 Minolta), and chroma (C) and hue (h°) values were calculated from these data according to McGuire (1992) using the following formulas:

$$C = \left(a^2 + b^2\right)^{1/2} \text{ and the hue angle } h^\circ = \arctan(b/a)$$

Cracking Index (%)

Harvesting for fruit cracking was performed between 08:00 and 10:00 in the morning. Fifty fruits without defects were selected and immediately immersed in 2-liter plastic containers filled with distilled water (20 ± 1 °C for 2, 4, and 6 h). Cracked fruits were counted and separated every 2 h. Cracked fruits were discarded from the water at each time interval. Cracked fruits were removed and counted, and fruits without cracks were reincubated. After 2, 4, and 6 h, the fruits were observed for macroscopic cracks and the cracking index (CI) was calculated according to the following formula: $CI = [(5a + 3b + c) \times 100]/250$, where a, b, and c represent the number of cracked fruits at 2, 4, and 6 h, respectively (Christensen 1972; Correia et al. 2020).

Determination of Total Antioxidant Activity

The radical-scavenging activity of 2,2-diphenyl-1-picrylhydrazyl (DPPH) was evaluated as described by Ganhão et al. (2019), with slight modifications. Briefly, 0.06 µM ethanolic DPPH was freshly prepared. Then, 1950 µl of DPPH was added to 50 µl of juice sample. The mixture was vortexed for 1 min. Measurements were calculated from the absorbance at a wavelength of 515 nm using a Multiskan™ GO microplate spectrophotometer (FI-01620 Vantaa, Finland). The radical-scavenging activity (percent DPPH inhibition) was calculated using the following equation:

$$\%\,\text{Inhibition} = 100\times [\text{Abs control} - (\text{Abs sample} - \text{Abs blank})]/[\text{Abs control}]$$

Where, Abs sample was the absorbance of the reaction in the presence of the sample (sample dilution ± DPPH solution), Abs blank was the absorbance of the blank for each sample dilution (sample dilution ± DPPH solvent), and Abs control was the absorbance of the control reaction (sample solvent ± DPPH solution).

Determination of Total Phenol Content

A 0.5-g sample of sweet cherry puree was weighed and 2.5 ml of 80% methanol was added. The mixture was vortexed for 1 min and then centrifuged at 4000 rpm for 4 min. With minor adjustments, the Folin–Ciocalteu method (Krawitzky et al. 2014) was used to calculate the total phenolic content. A UV/VIS spectrophotometer (Thermo Fisher Scientific, FI-01620 Vantaa, Finland) was used for the analysis. Briefly, 1 ml of the juice sample was mixed with 5 ml of 80% methanol. The mixture was centrifuged at 5500 rpm for 10 min and 250 µl of Folin–Ciocalteu reagent was added to 50 µl of the supernatant. Then 750 µl of 20% (w/v) Na_2CO_3 was added and incubated for 2 h at room temperature. The absorbance was then measured against a blank at 760 nm. Results are expressed as milligrams GAE/100 g fresh weight (FW) base. Results were determined using a calibration curve constructed daily using known amounts of gallic acid (GA) standards.

Table 1 Effects of treatments on morphological characteristics of cv. 'Royal Tiago' sweet cherry

	Control	AN	$CaCl_2$	GB	Si
Fruit weight	7.19 ± 0.15b	7.58 ± 0.3ab	7.85 ± 0.23ab	7.71 ± 0.12a	7.5 ± 0.07b
Seed weight	0.38 ± 0.7 ns	0.33 ± 0.01 ns	0.35 ± 0.03 ns	0.32 ± 0.01 ns	0.34 ± 0.02 ns
Fruit width	19.9 ± 1.2 ns	19.9 ± 0.1 ns	20.2 ± 0.4 ns	20.6 ± 0.3 ns	19.7 ± 0.1 ns
Fruit length	25.1 ± 1.7 ns	24.5 ± 0.3 ns	25.4 ± 0.6 ns	26.3 ± 0.1 ns	24.9 ± 0.2 ns
Fruit height	21.6 ± 1.4 ns	21.7 ± 0.5 ns	21.8 ± 0.4 ns	22.3 ± 0.3 ns	21.2 ± 0.7 ns
Stem length	22.4 ± 1.6 ns	20.9 ± 3.4 ns	23.8 ± 0.8 ns	26.1 ± 4 ns	20.4 ± 1.2 ns
L	23.7 ± 1.3b	28.1 ± 2.2ab	33.3 ± 4.7a	34.2 ± 2.5a	30.8 ± 0.9ab
a	22.9 ± 3.1b	22.6 ± 0.5b	22.4 ± 1.0b	28.2 ± 1.7a	26.5 ± 1.9ab
b	8.9 ± 0.9b	9.4 ± 0.5ab	8.8 ± 0.9b	11.7 ± 1.1a	11.5 ± 1.0a
Hue angle	21.7 ± 1.5ab	22.4 ± 0.6ab	21.0 ± 0.4b	23.0 ± 0.2b	23.7 ± 1.2a
Chroma	24.6 ± 3.11b	24.5 ± 0.7b	24.0 ± 1.3b	30.5 ± 1.9a	28.9 ± 2.0ab
Firmness	35.9 ± 2.4c	48.0 ± 0.7b	52.4 ± 0.9ab	56.6 ± 3.9a	54.4 ± 2.4a

Different letters in the same row indicate statistically significant differences at 0.05 level
ns non-significant, *AN Ascophyllum nodosum, GB* glycine betaine

Determination of Total Anthocyanin Content

The total anthocyanin (TA) concentration was calculated using the modified pH differential method (Cheng and Breen 1991).

Sugar and Organic Acids

For the analysis of sugar and organic acid components, homogenized samples were prepared and analyzed according to the high-performance liquid chromatography (HPLC) method developed by Bozan et al. (1997). One gram of sample was weighed and 4 ml of ultrapure water was added. After filtration, the samples were prepared for analysis. Both sugar and organic acid contents were determined by HPLC (Shimadzu, Prominence LC-20A) in three replicates. Organic acids were analyzed with a UV detector and a Transgenomic 87H (7.8 × 300 mm) column, while sugars were analyzed with a refractive index (RID) detector and a Coregel-87C (7.8 × 300 mm) column. Calibration curves for the reference standards were generated, and the content was determined based on these calibration curves.

Statistical Analyses

Results are expressed as mean ± standard error. Data were analyzed by analysis of variance (R version 4.4.1; R Foundation for Statistical Computing: $p \leq 0.05$, procedure aov), and multiple comparisons of treatment means were tested with Tukey's multiple range test (R version 4.4.1; R Foun-

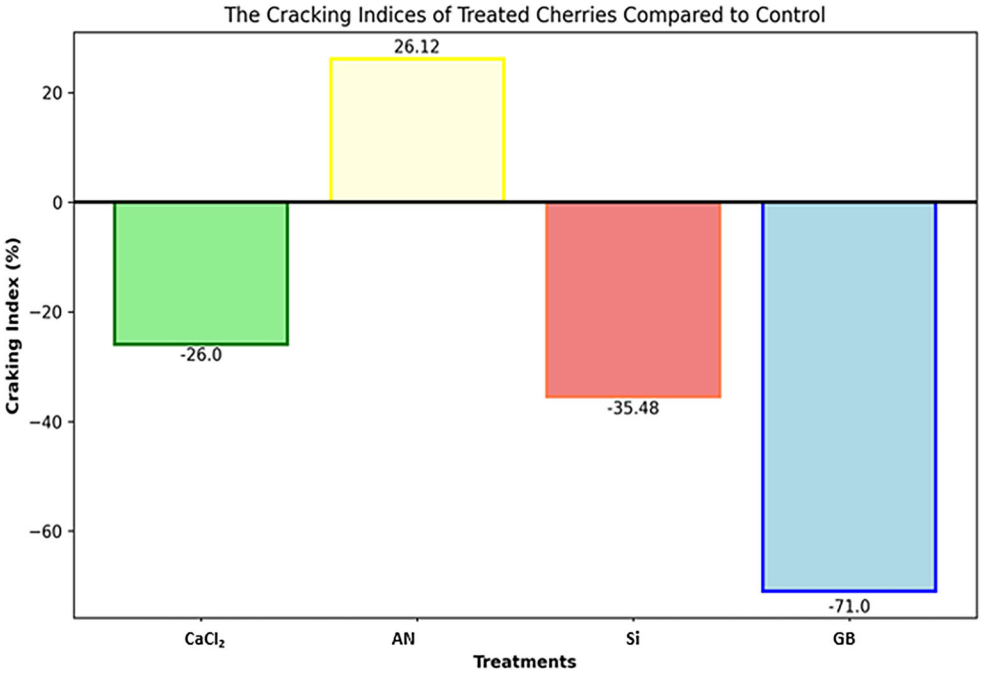


Fig. 1 Comparison of the cracking indices of sweet cherries under four different spray treatments with the control group. The values indicate the percentage (%) change in the cracking index relative to the control, highlighting either an increase or decrease for each treatment. *AN Ascophyllum nodosum*, *GB* glycine betaine

dation for Statistical Computing, Vienna, Austria: $p \leq 0.05$, package Agricola 1.3 7, procedure Duncan test). Using R Studio (version 4.4.1), principal component analysis was performed with the FactoMineR package (Kassambara 2017). To assess the strength and direction of linear relationships between variables, Pearson's correlation test was applied using the "rcorr" function from the Hmisc package in the R statistical program (R version 4.4.1).

Results

Fruit Dimensions, Color Indices, and Fruit Firmness

The effects of AN, Ca, GB, and Si treatments on the fruit morphological characteristics of 'Royal Tiago' sweet cherry are shown in Table 1. The analysis of seed weight (g), fruit width (mm), fruit length (mm), fruit height (mm), and stem length (mm) showed no significant differences among the treatments ($p < 0.05$; Table 1).

The analysis of the cracking index (CI) data (Fig. 1) showed significant differences between the different treatments ($p < 0.05$). Sweet cherries treated with GB showed the lowest CI values. When comparing the CI with the control group, there was a 26.12% increase in CI with the application of AN. On the contrary, the CI decreased by 26.00%, 35.48%, 44.59%, and 71.00% with Ca, Si, and GB treatments, respectively.

The exogenous spraying treatments had a profound effect on the differences in sugar components and organic acid contents. The highest contents of glucose, xylose, fructose, and total sugars were found in sweet cherries sprayed with $CaCI_2$ (Table 2). AN, $CaCI_2$, and GB increased the total sugar content and sugar components in sweet cherries compared to the control. In all spray treatments, glucose was identified as the highest sugar component, followed by fructose, sucrose, and xylose. The main organic acids determined in cv. 'Royal Tiago' were malic acid followed by tartaric acid, succinic acid, citric acid, and oxalic acid. The organic acids were influenced by the all-foliar treatments ($p < 0.05$; Table 2). Compared to the control, the levels of malic, tartaric, citric, succinic, and oxalic acids were highest in the $CaCI_2$ treatment, followed by the GB, AN and Si treatments. This indicates that the $CaCI_2$ treatment resulted in the most significant increase in these organic acids.

The results showed large variations in biochemical compounds and bioactive contents among the treatments (Table 3). The GB, Si, and Ca treatments had similar total phenolic contents, which were significantly higher than those of the control and AN treatment. Notable differences in total antioxidant activity were observed among the treatments. The GB treatment had the highest total antioxidant content, while the control had the lowest compared to the other spray treatments. Total anthocyanin content reached its highest value in sweet cherries treated with GB. There were no significant differences among the control, AN, and Si treatments with respect to total anthocyanin (Table 3).

All treatments resulted in a decrease in the Brix levels of sweet cherries (Table 3). The treatments did not cause any significant changes in the pH levels of the sweet cherries. In terms of acidity (%), AN, Ca, and Si treatments showed lower values.

Principle Component Analyses and Pearson Correlation Analysis

The results of principal component analysis (PCA), including the corresponding eigenvalues and matrix components, are shown in Fig. 2. The biplot represents the first two principal components (PC1 and PC2), which explain 31.9% and 29.0% of the total variance, respectively. The vectors represent the original variables, with the length of each vector, such as xylose, fructose, glucose, total sugars, tartaric acid, and DPPH, indicating the strength of the variable's contribution to the principal components.

The angle between the vectors reflects the correlation between the variables; smaller angles indicate positive cor-

Table 2 Effects of treatments on organic acids and specific sugar content of cv. 'Royal Tiago' sweet cherry

	Control	AN	$CaCI_2$	GB	Si
Sucrose	426 ± 81c	681 ± 5a	568 ± 8b	700 ± 1a	461 ± 4c
Glucose	3539 ± 20c	3931 ± 2b	4272 ± 9a	3542 ± 8c	2784 ± 9d
Xylose	116 ± 4b	118 ± 6b	140 ± 8a	92 ± 2c	92 ± 4c
Fructose	3103 ± 10b	3512 ± 13a	3726 ± 17a	3063 ± 6b	2407 ± 7c
Total sugar	7185 ± 17b	8242 ± 19a	8706 ± 19a	7398 ± 11b	5744 ± 9c
Oxalic acid	12.1 ± 1b	10.3 ± 1b	20.5 ± 3a	11.1 ± 1b	10.6 ± 1b
Citric acid	99 ± 2d	140 ± 2a	106 ± 7cd	129 ± 6ab	117 ± 5bc
Tartaric acid	423 ± 13b	499 ± 16a	490 ± 15a	509 ± 16a	397 ± 8b
Malic acid	335 ± 2d	359 ± 3c	515 ± 9a	337 ± 8d	414 ± 5b
Succinic acid	159 ± 12bc	177 ± 8ab	189 ± 12a	189 ± 13a	150 ± 9c

Different letters in same row indicate statistically significant differences at 0.05 level
ns non-significant, *AN Ascophyllum nodosum, GB* glycine betaine

Table 3 Effects of treatments on biochemical and bioactive content of cv. 'Royal Tiago' sweet cherry

	Control	AN	$CaCl_2$	GB	Si
Total phenol	3731 ± 165c	4342 ± 445c	5000 ± 400 ab	5394 ± 248a	4857 ± 119ab
DPPH	64.2 ± 1.2d	68.2 ± 1.9bc	71.0 ± 1.0b	74.4 ± 1.2a	66.7 ± 1.1cd
Anthocyanin	6.10 ± 0.1b	6.22 ± 0.4b	6.02 ± 0.3b	9.02 ± 0.2a	5.78 ± 0.07b
Brix°	16.0 ± 0.1a	13.2 ± 0.2bc	14.2 ± 0.4b	14.0 ± 0.4bc	13.7 ± 0.1c
pH	3.15 ± 0.03 ns	3.19 ± 0.03 ns	3.22 ± 0.04 ns	3.21 ± 0.03 ns	3.25 ± 0.12 ns
Acidity	8.23 ± 0.2a	7.21 ± 0.5b	7.18 ± 0.12b	8.7 ± 0.13a	7.89 ± 0.6ab

Different letters in same row indicate statistically significant differences at 0.05 level
ns non-significant, *AN Ascophyllum nodosum, GB* glycine betaine

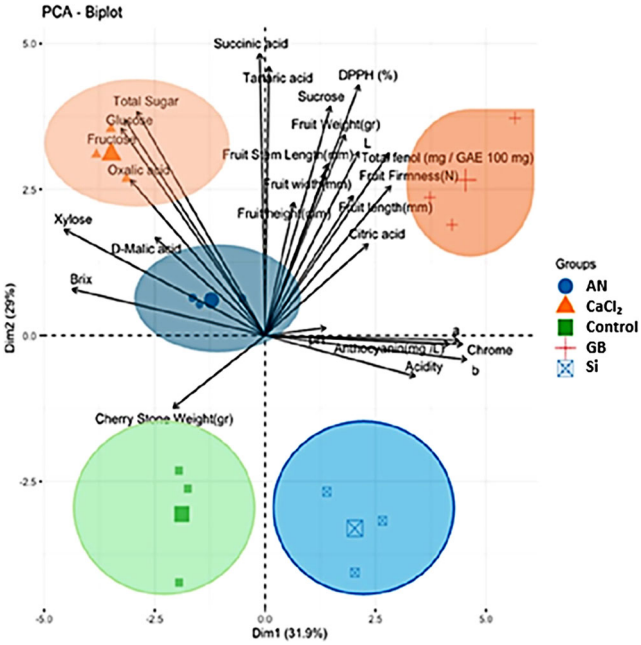


Fig. 2 Principal component analysis for morphological parameters and biochemical contents of the 'Royal Tiago' sweet cherry cultivar under five different treatments. *AN Ascophyllum nodosum*, *GB* glycine betaine

relations. Conversely, larger angles (close to 180°) indicate negative correlations (Fig. 2).

This analysis highlights the primary relationships and differences between the samples in the dataset. Notably, five different treatments are clearly separated, indicating that samples within each group share similar characteristics (e.g., $CaCl_2$ and AN) while being significantly different from those in other groups. In addition, GB showed significant positive correlations with DPPH, firmness, total phenol, and sucrose content, but significant negative correlations with cracking index (Fig. 2).

Pearson's correlation coefficient was calculated to assess the contributions of all morphological and biochemical parameters (Fig. 3).

A strong negative correlation was observed between the cracking index (%) and total phenolic content (r = –0.75) as well as with DPPH antioxidant activity (r = –0.69) and color parameters (L, a, b; r = –0.64, r = –0.67, r = –0.58 respectively, $p < 0.05$) and fruit firmness (r = –0.61). Furthermore, a significant negative correlation was observed between the cracking index and acidity (r = –0.57, $p < 0.05$). Furthermore, a significant negative correlation ($p < 0.05$) was found between redness (a) and glucose (r = –0.53), xylose (r = –0.69), and fructose (r = –0.56). A strong correlation was also observed between Brix and the content of glucose (r = –0.53), xylose (r = –0.84) and fructose (r = –0.53).

Discussion

The cracking index is a notable parameter in the effectiveness of all treatments. All spray treatments, except AN, provided a significant reduction in cracking index (%) compared to the control. Lahdenpera (2006) demonstrated that GB treatment reduced fruit cracking in 'Garnet' sweet cherries. Furthermore, Li et al. (2019) suggested that GB was an effective method to improve fruit firmness in sweet cherries when applied multiple times and earlier. Another promising approach to mitigate sweet cherry cracking is Si and Ca spray treatment. A reduction in the cracking index of GB-treated sweet cherry 'Sweetheart' was reported by Correia et al. (2020). A previous study reported an approximately 50% reduction in cracking index (CI) when Ca was applied to 'Sweetheart' (Michailidis et al. 2021). Ca treatments also reduced the cracking index (%) in sweet cherry cultivars 'Sweetheart' and 'Skeena' (Correia et al. 2020). In addition to the focus on Ca in mitigating fruit cracking, Si spray treatments were found to be effective in citrus (Wang et al. 2024) and lychee (Huang et al. 2001). Two hypothetical mechanisms have been proposed to explain the reduction of fruit cracking by calcium. According to several authors (Bastos et al. 2015; Usenik et al. 2015), the first mechanism suggests that calcium promotes crosslinking between the cellular components of the cell wall, modifying the mechanical properties of both the fruit skin and flesh, making them more rigid. The second mechanism proposes that the reduction in cracking is due to reduced water uptake. The application of calcium and potassium salts creates a more negative osmotic potential in the external water, which re-

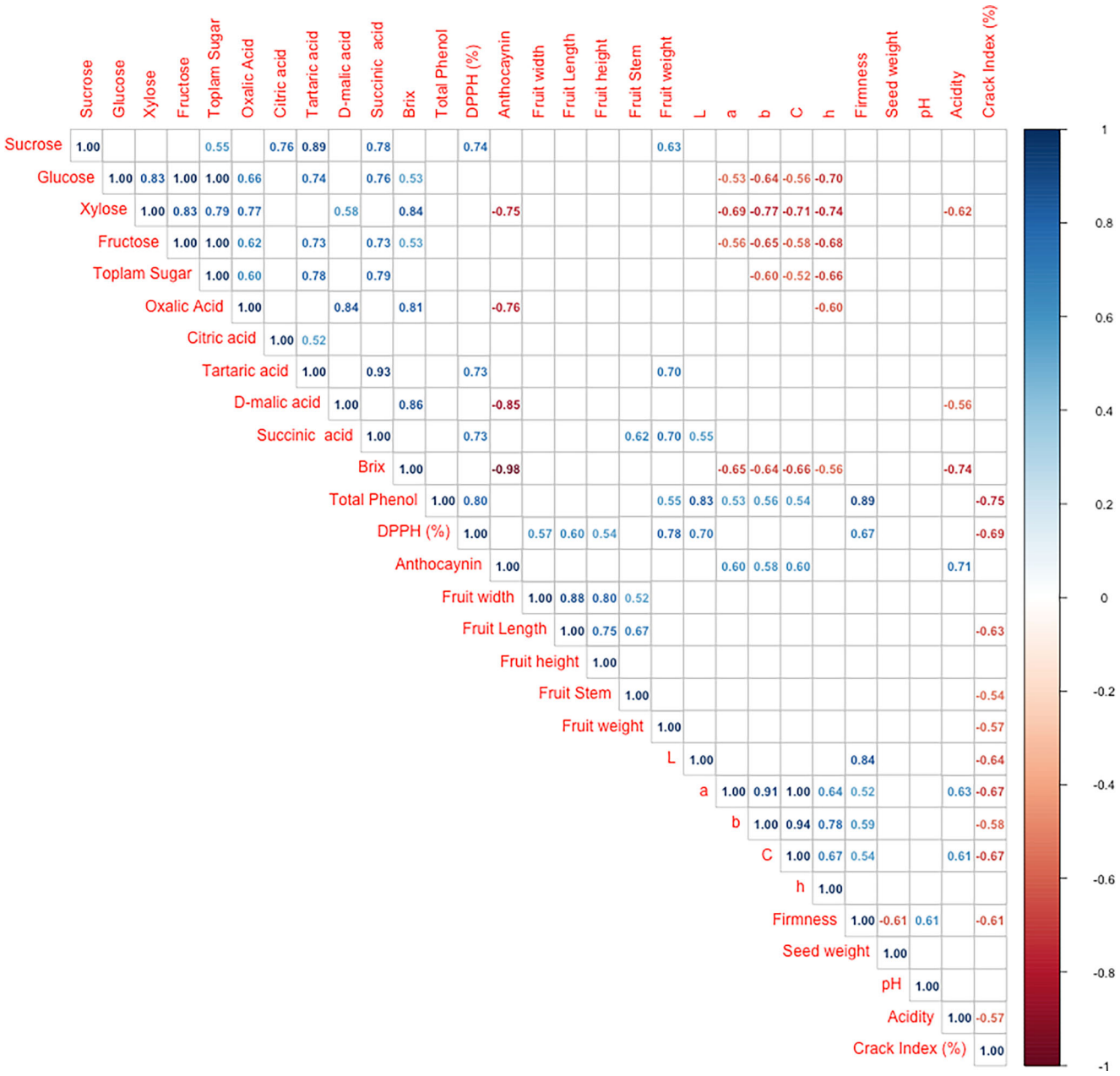


Fig. 3 Pearson correlation analysis for morphological parameters and biochemical and bioactive contents of the 'Royal Tiago' cherry cultivar under five different treatments. Only significant correlations ($p < 0.05$) are displayed

duces water uptake by the fruit. This lower water uptake rate ultimately contributes to the observed reduction in cracking incidence (Wójcik et al. 2013, Saran et al. 2015). Based on our results, the reduction in cracking observed after foliar treatments can be attributed to the second hypothesis. Correia et al. (2020) suggest that during the early stages of fruit development, the thickness of the cuticle plays a key role in limiting the cracking index, as only a significant correlation with histological parameters was found. In contrast, at later stages of fruit development, larger epidermis, hypodermis, and parenchyma cells appear to reduce the cracking index (CI). Kertesz and Nebel (1935) noted that cultivars more tolerant to cracking tended to have larger hypodermis cell sizes. Correia et al. (2020) demonstrated that Paβ-Gal plays an important role in conferring cracking tolerance, as higher levels of Paβ-Gal transcripts were found in fruits treated with AN + Ca and GB + Ca at the early stage of fruit development and in Ca-treated fruits at the late stage. However, they did not observe a high cracking index in AN + Ca-treated fruits, and increased expression of Paβ-Gal was not detected in ABA + Ca-treated fruits.

Fruit firmness (FF) is a critical quality factor in sweet cherries that is associated with improved resistance to decay and mechanical damage, ultimately extending shelf life (Barreit and Gonzales 1994). Interestingly, sweet cherries sprayed with AN, Ca, GB, and Si increased firmness at harvest. Li et al. (2019) found that GB-treated 'Regina' sweet cherries had significantly higher calcium content, which could be attributed to the increased fruit firmness. Furthermore, Gonçalves et al. (2020) found the same results in cv. 'Staccato' sweet cherries. These results are similar to ours. Moreover, exogenous Si treatment increased fruit firmness in apple (Karagiannis et al. 2021) and tomato fruit (Costan et al. 2020), suggesting that foliar Si is efficient in promoting greater firmness retention. Glazowska et al. (2018) reported that foliar Si enhanced cellulose and hemicellulose deposition in rice, highlighting the role of Si in cell wall formation and softening processes. The increase in fruit firmness observed in Si-treated sweet cherries suggests that Si may play a critical role in stabilizing the cell wall by protecting it from degradative enzymes such as polygalacturonase and pectate lyase. The current understanding of the role of silicon (Si) in fruit firmness remains limited, highlighting the need for further research, particularly regarding the potential effects of Si treatments on postharvest metabolism and ripening processes in sweet cherries. Dong et al. (2019) demonstrated that higher calcium content in fruit tissues contributed to firmer sweet cherries. To significantly influence fruit firmness (FF), it is necessary to increase tissue calcium (Ca) levels to 800–1000 mg/kg dry weight (DW). However, Ca levels above 1000 mg/kg DW can result in fruit surface damage (Sams and Conway 1993). Calcium treatment consistently results in increased fruit firmness (FF) and higher tissue calcium levels. This may increase resistance to postharvest disorders without negatively affecting skin color development (Tsantili et al. 2007; Wójcik et al. 2013). However, the fruit's response to calcium treatments varies with cultivar and calcium source as well as with the frequency, timing, and concentration of treatment (Wójcik et al. 2013; Michailidis et al. 2021).

Color is one of the most important characteristics used by buyers and consumers to determine the maturity and ripeness of fruit (Ercisli et al. 2005; Fidan et al. 2020; Urun et al. 2021; Haghshenas et al. 2024; Imrak et al. 2024; Rymbai et al. 2024; Zor 2024). Sweet cherry cultivars vary in their optimal harvest color; however, deep-red-colored sweet cherries are generally preferred because they are associated with sweetness, flavor, and high nutritional value (Peris-Felipo et al. 2020). In general, the results of the preharvest spray treatments had the same significant effect ($p < 0.05$) on L, a, b, C, and the color angle of sweet cherries. Four foliar treatments increased the values of color parameters compared to the control; however, these treatments showed statistically similar values of color parameters in sweet cherry cultivar 'Royal Tiago.' Results regarding the effect of Si and GB on color parameter dissimilarities during fruit ripening are both inconsistent and limited (Jayawardana et al. 2014; González-Terán et al. 2020; Basile et al. 2021). For example, Karagiannis et al. (2021) found that color parameters such as hue angle were reduced by Si application in apple. However, Peris-Felipo et al. (2020) found no significant effect of Si on the color of strawberries when investigating its effect on quality parameters. Correia et al. (2020) found that GB may result in a darker red color in sweet cherry cultivars 'Skeena' and 'Sweetheart.' In contrast, Li et al. (2019) found that sweet cherry cultivars 'Lapins' and 'Regina' were not affected by GB. Therefore, it is difficult to draw definitive conclusions regarding the effects of GB and Si on fruit color development.

Our results indicate that except for fruit weight, growth parameters (including fruit height, width, length, and stem length) and ripening characteristics such as pH were not affected by the spray treatments. In developing strategies to increase fruit weight, spray treatments such as AN, GB, Ca, and Si are of great importance ($p < 0.05$). In previous studies, Santos et al. (2023) found that sweet cherries sprayed with AN had slightly increased fruit weight and larger diameter, while Ca decreased fruit weight and similar diameter values. Similar results were obtained by Correia et al. (2015), where AN-sprayed sweet cherry cultivars 'Sweetheart' and 'Skeena' resulted in increased fruit dimensions such as weight and diameter. Correia et al. (2018) suggested that both calcium and glycine betaine (GB) spray treatments could increase sweet cherry fruit weight, although no significant effect on fruit firmness was observed. Treatment with AN in cv. 'Staccato' also resulted in larger fruits and similar weight and diameter results compared to the control (Gonçalves et al. 2020). In addition, biostimulant treatment increased fruit diameter in sweet cherry cultivars 'Kordia' and 'Regina' (Gonçalves et al. 2020). Biostimulants such as AN provide a promising new approach to stimulating plant growth and increasing yield, as reported by Basak (2008) and Colavita et al. (2011). However, in other studies, no clear effect of Si on fruit weight was seen for apple (Karagiannis et al. 2021), cucumber (González-Terán et al. 2020), or pepper (Jayawardana et al. 2014). Therefore, it is difficult to draw definitive conclusions regarding the effects of Si on fruit weight.

All spray treatments had a significant effect on Brix and acidity of sweet cherry fruit. AN, Ca, GB, and Si treatments significantly decreased the concentration of Brix and acidity. Gonçalves et al. (2020) stated that sweet cherries treated with GB and AN had the highest Brix and pH and the lowest acidity values. Abou-Aly and Mady (2014) observed that glycine betaine (GB) increased total carbohydrates, vitamin C, and total soluble solids (TSS) in ripened sweet pep-

per (*Capsicum annuum* L.) fruits grown under high-temperature conditions. Similarly, de Melo et al. (2018) reported higher TSS and lower titratable acidity (TA) in mango fruits after external treatment with *Ascophyllum nodosum* extract. In addition, De Souza et al. (2011) found a decrease in the acidity of mangoes following similar treatments. They also found a reduction in the acidity of mango fruits. *Ascophyllum nodosum* has been shown to interfere with normal acid metabolism during respiration, resulting in delayed senescence and effectively extending the shelf life of mangoes. Although not the focus of our study, sweet cherries may also benefit from similar effects, potentially extending their availability to consumers (Pereira et al. 2020). Similarly, Correia et al. (2015) found that *Ascophyllum nodosum* treatment significantly increased fruit length, diameter, and pH in a study of sweet cherries from the 'Skeena' and 'Sweetheart' cultivars, which is consistent with our observations in the current study for cherries from the 'Royal Tiago' cultivar.

Total phenolic content showed a significant inverse relationship (r = –0.75), as did DPPH antioxidant activity (r = –0.69). Negative correlations were also observed with color parameters, including L (lightness; (r = –0.64), a (red-green; r = –0.67), and b (yellow-blue; r = –0.58), and with fruit firmness (r = –0.61). Fruit firmness, along with lower soluble solids and acidity, could reduce the susceptibility of fruit to cracking (Pereira et al. 2020). We found a strong negative correlation between the cracking index (%) and several key parameters. The average values for DPPH radical scavenging and total anthocyanin content were highest in cherries sprayed with GB. These results are in agreement with Gonçalves et al. (2020), who found that DPPH and polyphenol content were significantly affected by GB spray treatment. Several studies have reported beneficial effects of AN treatments associated with increases in plant growth, yield parameters, and improvements in protein and nutrient content (Sharma et al. 2017; Layek et al. 2018). Frioni et al. (2018) highlight the positive effect of AN extract on phenolic and anthocyanin levels in grapevine cultivars used for red wine production. These results are consistent with observations from other horticultural crops, which also reported increases in anthocyanin and phenolic content (Roussos et al. 2009; Lola-Luz et al. 2013; Fan et al. 2011). These effects could be attributed to the composition of AN, which contains organic and inorganic compounds, including unique and complex polysaccharides, plant hormones (Sivasankari et al. 2006; Khan et al. 2009), and other hormone-like compounds such as sterols and polyamines (Craigie 2011). By measuring the content of fructose, glucose, sucrose, xylose, and total sugars in the sweet cherries, it was found that there were differences among all the spray treatments, with the fructose, glucose, and total sugar content of the cherries sprayed with $CaCl_2$ being the highest. Meanwhile, glucose was the most abundant primary metabolite of sweet cherry fruit, followed by fructose, sucrose, and xylose. Sweet cherries treated with AN, GB, and Si showed an increase in sucrose content, while fructose content remained relatively stable. By examining the organic acid content of sweet cherries, it was found that Ca and Si significantly increased the malic acid content. AN and GB significantly increased the content of citric, succinic, and tartaric acids. Karagiannis et al. (2021) found that sugar and organic acid contents were significantly increased in apple by Si treatment. These results may be related to the increased photosynthetic activity observed in apple leaves after foliar application of Si (Karagiannis et al. 2021), a phenomenon also observed in other plants (Song et al. 2014), which ultimately leads to sugar accumulation. Malate, which is regulated by the enzymatic activity of malate dehydrogenase (MDH) and phosphoenolpyruvate carboxylase (PEPC) through the phosphoenolpyruvate pathway, serves as an essential carbon source in various metabolic processes, including gluconeogenesis, respiration, and the TCA cycle. The accumulation of malic acid observed in Ca- and Si-treated sweet cherries suggests that Si and Ca supplementation may directly influence MDH and PEPC biosynthesis, potentially regulating key traits in sweet cherries.

Conclusion

Preharvest $CaCl_2$, GB, and Si foliar treatments were more effective in increasing yield and reducing the incidence of fruit cracking in sweet cherry 'Royal Tiago.' Therefore, these foliar sprays are attractive compounds for improving the physiological performance and yield of sweet cherry trees and may represent a promising approach to mitigate cherry cracking. Foliar treatments with AN, GB, and Ca significantly increased sugar components and malic acid content, indicating their potential to improve the quality and biochemical composition of treated trees. Although glycine betaine (GB) and silicon (Si) may become standard cultural practices in the near future, further research is needed to understand the effects of environmental conditions, concentration levels, and timing of exogenous foliar applications. These studies will be essential for the development of new cultivars and specific management strategies.

Acknowledgements The author extends his appreciation to Cukurova University.

Author Contribution B. Imrak: conceptualization, methodology, software, validation, formal analysis, investigation, resources, data curation, writing—original draft preparation, writing—review and editing, visualization, supervision.

Declarations

Conflict of interest B. Imrak declares that he has no competing interests.

Ethical standards For this article no studies with human participants or animals were performed by any of the authors. All studies mentioned were in accordance with the ethical standards indicated in each case.

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