Horticulture, Environment, and Biotechnology (2023) 64:371–378
https://doi.org/10.1007/s13580-022-00486-8

RESEARCH REPORT

Canopy-applied silicon is an effective strategy for reducing sweet cherry cracking

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Received: 9 May 2022 / Revised: 14 September 2022 / Accepted: 14 September 2022 / Published online: 30 March 2023
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Abstract
Fruit cracking caused by rainfall prior to harvest, a major problem in sweet cherry production, is being exacerbated by climate change. Currently, pre-harvest spraying with calcium salt solutions is the prevalent technique to reduce fruit cracking in cherry orchards not covered by plastic roofs. This study evaluated the effectiveness of canopy-applied silicon in the reduction of sweet cherry cracking under different field conditions. Four field trials were conducted on mature trees of the cultivars Van, New Star, and Emperor Francis. Treatments included water (control), calcium chloride, and sodium silicate. Multiple sprays (three) were applied weekly from fruit onset of color to approximately 1 week before harvest. The results showed that under conditions conducive to cracking, sodium silicate reduced the percentage of cracked fruits to a similar or larger extent than calcium chloride. This study highlights how canopy-applied silicon sources may effectively contribute to reducing cherry cracking, acting as an alternative technique to other preventive methods.

Keywords Cell wall extensibility · Cuticle · Cuticular fractures · cryo-SEM · Energy-dispersive X-ray microanalysis

1 Introduction

Fruit cracking is a physiological disorder that can dramatically depreciate cherry fruits, causing heavy economic losses starting from the fruit's onset of color (Sekse 1995; Sekse et al. 2005; Simon 2006; Knoche and Winkler 2017; Correia et al. 2018). Recently, climate change has intensified the occurrence and damages caused by rain-induced cracking in sweet cherry and other important horticultural crops. Fruits are particularly vulnerable to cracking from onset of color to maturity, mostly in conjunction with high atmospheric humidity or rainfall events (Christensen 1996).

The harvest value is extremely compromised when the incidence of cracking is higher than 20–30% (Hansen and Proebsting 1996). This is recurrent for susceptible genotypes, such as cv. Van, where cracking can reach almost 100% of the marketable fruits. Moreover, lesions localized on the drupe represent a preferential access route to the spread of pathogens such as *Monilinia laxa* and *Botrytis cinerea* (Børve et al. 2000), causing additional losses.

Many hypotheses have been proposed to explain the phenomenon of fruit cracking, even though the responsible mechanisms are not completely elucidated (Correia et al. 2018).

Communicated by Heakeun Yun.

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The incidence and severity of cherry cracking are influenced by genotypic, environmental, and agronomic factors (Correia et al. 2018). The susceptibility to cracking differs largely among cherry cultivars (Sekse 1995; Christensen 1996; Demirsoy and Demirsoy 2004; Measham et al. 2009; Correia et al. 2018) and can be modulated by the rootstock (Cline et al. 1995; Sekse 1995; Simon et al. 2004). In addition, the fruit cuticle plays a crucial role in cracking susceptibility (Martin and Rose 2014; Tafolla-Arellano et al. 2018; Wang et al. 2021).

The reduction of cherry cracking often involves the use of physical-mechanical systems (plastic roofs, helicopters, or air blast blowers), chemicals (calcium-based compounds, mineral sprays, antitranspirants, and growth regulators), or biostimulants (e.g., seaweed extracts) (Sekse 1995; Sekse et al. 2005; Simon 2006; Knoche and Winkler 2017; Correia et al. 2018). Plastic roofs reduce cracking damage but are costly (Simon 2006; Correia et al. 2018).

Pre-harvest calcium sprays generally reduce sweet cherry cracking (Demirsoy and Bilgener 1998; Sekse 1998), as calcium plays a critical role in maintaining the structural integrity and firmness of the cell wall of the fruits (Christensen 1972; Sekse 1995). However, calcium sprays can reduce fruit size (Looney 1986; Facteau et al. 1987).

The techniques used to reduce fruit cracking are hardly effective when its incidence is high (e.g. from frequent rainfall) and present some drawbacks. Consequently, it is necessary to develop agronomic strategies that prevent the final appearance of macrocracks, without generating adverse impacts on the fruits and the tree. Modifications of cell wall properties (extensibility and stability) of the epidermis and the preservation of fruit cuticles by agronomic means are likely to contribute to reduced cracking.

Silicon (Si) is a beneficial element whose positive effects are mainly associated with its high deposition in plant tissues, enhancing their strength and rigidity (Ma and Takahashi 2002; Ma and Yamaji 2006). Si is capable of increasing cell wall extensibility and thereby the elongation process in the meristematic zone (Hossain et al. 2002; Hattori et al. 2003; Bat-Erdene et al. 2021). In addition, Si forms a protective layer that prevents the penetration of water and fungal pathogens and stimulates defense reactions against pathogens (Ma and Takahashi 2002; Ma and Yamaji 2006; Bat-Erdene et al. 2021).

The objective of this study is to evaluate the effectiveness of canopy-applied Si sources in the reduction of sweet cherry cracking through a range of field experiments under different environmental conditions.

Table 1 Atmospheric average temperature (°C), relative humidity (%), and accumulated precipitation in the period between onset of color and harvest for each experiment

	Start date of onset of color	Harvest date	Average temperature (°C)	Average relative humidity (%)	Accumulated precipitation (mm)
Experiment 1	May 17, 2002	June 13, 2002	19.6	71	9
Experiment 2	June 27, 2003	July 14, 2003	21.2	72	42
Experiment 3	May 18, 2005	June 20, 2005	19.8	57	6
Experiment 4	May 7, 2007	June 04, 2007	20.7	58	37

2 Materials and methods

2.1 Plant material and growing conditions

Sweet cherry (*Prunus avium*) mature trees of cultivars Van, Emperor Francis, and New Star were subjected to canopy-applied treatment in four experimental sites, of which three were located in Northern Italy and one in Upper Midwestern, United States. Meteorological data for the period between onset of color and harvest are presented in Table 1.

Experiment 1 was conducted in 2002 in a mature cherry orchard located in Savignano sul Panaro (Modena, Italy) (44°29'N 11°02'E; 217 m a.s.l.) on plants of the cv. Van grafted on Colt.

Experiment 2 was conducted in 2003 in a mature cherry orchard located at the Clarksville Research Center, Michigan State University (Clarksville, Michigan, USA) (42°52'N 85°15'W; 251 m a.s.l.) on plants of the cv. Emperor Francis grafted on a cherry seedling.

Experiment 3 was carried out in 2005 in Vignola (Modena, Italy), (44°28'N 11°00'E; 125 m a.s.l.), in a mature orchard, on plants of cv. Van grafted on a cherry seedlings.

Experiment 4 was carried out in 2007 in Castelfranco Emilia (Modena, Italy), (44°34'N 11°05'E; 35 m a.s.l.) in a mature orchard, on plants of cv. New Star grafted on a cherry seedling.

2.2 Canopy-applied calcium and silicon treatments

Field experiments were conducted, during one growing season, in a completely randomized block design with three replicates. The following treatments were compared: (1) control (distilled water), (2) calcium chloride ($CaCl_2$) (5 g L^{-1}), and (3) sodium silicate ($Na_2OSiO_2)_3$ (36–40°Bé) (2.3 g L^{-1}). Each treatment was applied to a single branch within a tree. The pH of all solutions was adjusted to 5.5 using HCl. Solutions were applied in the late afternoon to the canopy through pressurized nebulizers, until the complete dripping of fruits and leaves.

Three (Experiments 1–3) and four (Experiment 4) randomly selected trees were sprayed for each treatment and

separated by at least two untreated trees along consecutive plots. Treatments were performed weekly from the onset of color until commercial harvest, for a total of three applications.

2.3 Sampling and measurement methods

The effects of treatments were evaluated at harvest. The percentage of cracked fruits was calculated based on the total number of treated fruits. In Experiments 1 and 2, the types of cracks (peduncular, dorsal, and apical end) were also assessed.

2.4 Fruit quality parameters

Fruit quality was evaluated, for all experiments, by fruit weight and soluble solids content (TSS) (Atago Digital Refractometer, Optolab, Modena, Italy). Fruit firmness (Effegi FT 011, tip ø 5 mm, Ravenna, Italy) (Experiments 1 and 4), pH, and juice titratable acidity (Experiment 4) (Crison Titromatic 1 S, Barcelona, Spain) were also determined.

Cherry skin color parameters were determined (Experiment 1) via a chromameter (CR-200 Chromameter, Minolta Co. Ltd., Osaka, Japan) using the digital representation model HLS (hue, lightness, and saturation). The hue axis (H) includes red at 0° and 360°. The brightness axis (L) ranges from 0 (black) to 100 (white). The saturation axis (S) ranges from 0 (darker color) to 100 (more intense color).

2.5 Silicon localization by cryo-scanning electron microscopy and energy-dispersive X-ray microanalysis

Analysis of the deposition of Si on the surface of the fruits and its possible penetration into the internal tissues was performed on the frozen-hydrated, whole, and freeze-fractured samples of cvs. Van (Experiment 3) and New Star (Experiment 4) by cryo-scanning electron microscopy (Cryo-SEM) and energy-dispersive X-ray microanalysis (McCully et al. 2009, 2010; Minnocci et al. 2018). At harvest, a portion of the surface and a parallelepiped of the regular Sect. (5 × 5 mm) from the epidermis until the seed of unwashed fruits (without stalk) were sampled. The fruit portions were immediately cryo-fixed by immersion in liquid nitrogen (−196 °C) and stored until Cryo-SEM analysis. For the internal tissue analysis, the frozen-hydrated (FH) samples were first mounted under liquid nitrogen gas in an aluminum stub with Tissue-Tek, freeze-fractured inside the liquid nitrogen to expose the internal texture, transferred to a dedicated cryo-preparation chamber (SEM Cryo Unit, SCU 020, Bal-Tech, Balzers, Liechtenstein), surface etched for 3 min at −80 °C under high vacuum ($P < 2 \times 10^{-4}$ Pa), and sputter-coated with 8-nm gold in an argon atmosphere ($P < 2.2 \times 10^{-2}$ Pa) to produce an electrically conductive surface. FH specimens were finally transferred to the cryo-stage (−180 °C) inside a scanning electron microscope (Philips SEM 515, Eindhoven, the Netherlands). Energy-dispersive X-ray microanalysis of FH specimens was performed with SEM using an acceleration voltage of 17 kV, a take-off angle of 16.5°, and a working distance (sample to the final lens of the SEM instrument) of 12.0 mm. Spectra from 0 to 20 keV were acquired for 120 s (live time) with a dead time of less than 20%, and collected at increments of 10 eV per channel with the electron beam focused on a spot area (diameter 40 nm) in the center of the selected cells (Fig. 1D). The background and element-specific peak spectra were analyzed using the program EDAX DX-4 2.0 (EDAX, San Francisco, CA), which fully deconvolutes the spectra and allows corrections for interference between elements (Servili et al. 2008).

2.6 Statistical analysis

Data were subjected to analysis of variance, and the comparison between treatments was performed by the Student Newman Keuls (SNK) test ($P \leq 0.05$).

3 Results

3.1 Effects of canopy-applied sodium silicate and calcium chloride on sweet cherry cracking

3.1.1 Experiment 1

The application of Si and $CaCl_2$ reduced to a similar extent the incidence of cracked fruits compared to water-sprayed fruits (Table 2). Most fruits showed cracks in the apical portion, and the type of cracking was not affected by treatments (Table 2). The application of sodium silicate to the canopy decreased the average weight of the fruit at harvest in comparison to control plants (Table 3), whereas fruit firmness was not affected by treatments (Table 3). Si treatment reduced hue, brightness, and saturation (Table 4).

3.1.2 Experiment 2

The highest percentage of cracked fruits was recorded in control plants. This value was reduced, with similar effectiveness, in the branches treated with sodium silicate and calcium salt (Table 2). In addition, differences were observed also in the type of cracking (Table 2). The supply of calcium chloride to the canopy reduced the average weight of the fruit and the soluble solids, while the sodium silicate increased the latter parameter (Table 3).

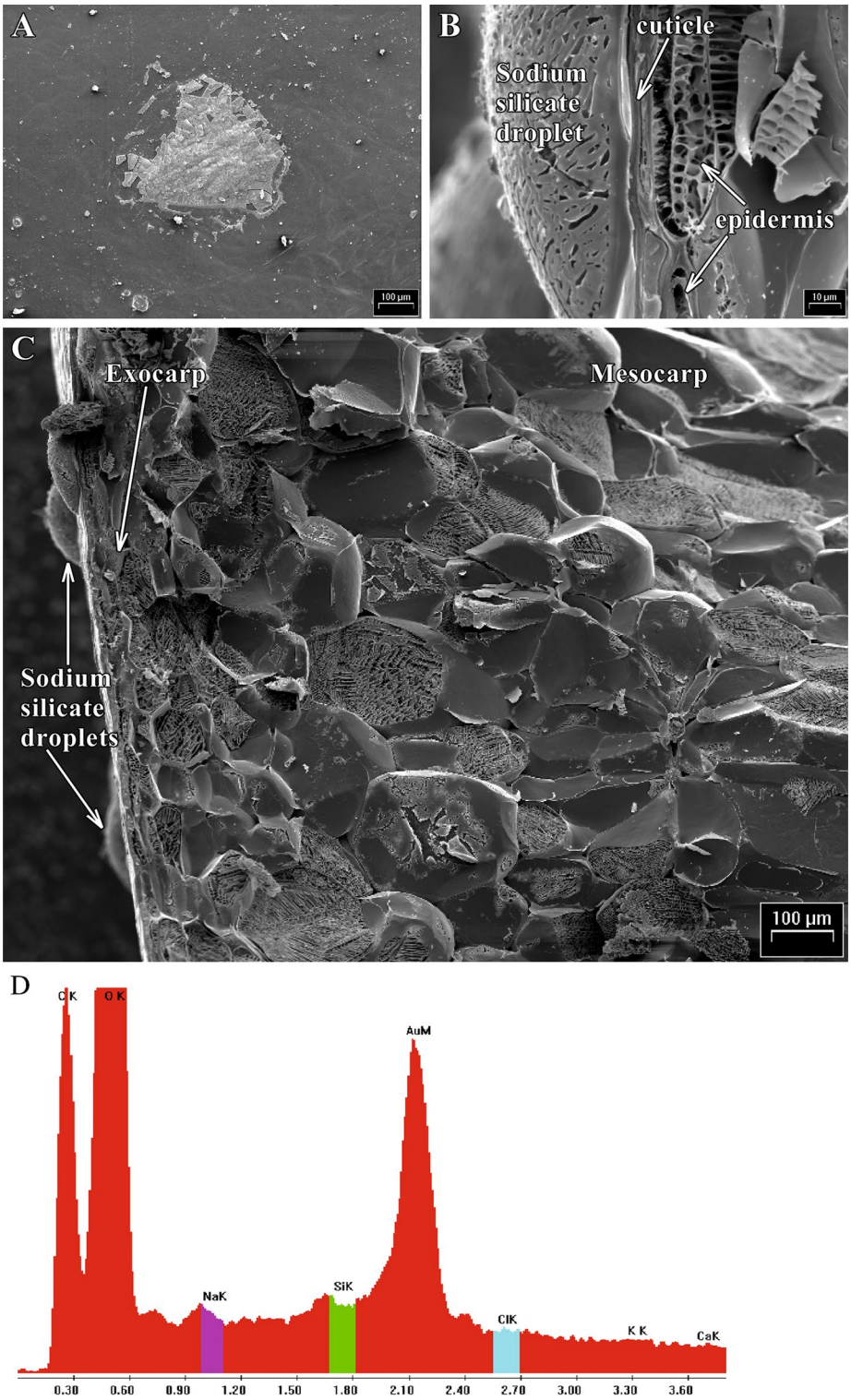

Fig. 1 (A): cv. Van, sodium silicate residue on the outer surface of the treated fruit; (B): cv. New Star, transverse freeze-fracture of exocarp in correspondence of a residual drop of sodium silicate; (C) the same plane of fracture of (B) at a lower magnification to display the different cell types examined with X-ray microanalysis; and (D) an X-ray spectrum representative of those detected both in cells of the epidermis and in the mesocarp, with the silicon content (SiK) highlighted in green

3.1.3 Experiment 3

The low rainfall that occurred during the experiment (Table 1) prevented the occurrence of cracking. Negligible cracking incidence was observed (less than 0.5%; data not shown), and it was not influenced by treatment. Similarly, treatments did not affect fruit weight at harvest (Table 3). Nevertheless, total soluble solids were enhanced by canopy-applied sodium silicate (Table 3).

Table 2 Effects of treatments on the incidence and type of fruit cracking (data expressed as a percentage)

		Fruit Cracking (%)				
	Treatments	Total	Apical	Peduncular	Dorsal	Multiple
Experiment 1	Control	27.9 a[1]	58	36	6	-
	Calcium chloride	13.4 b	74	26	0	-
	Sodium silicate	13.3 b	58	42	0	-
	Significance	*[2]	*n.s.*	*n.s.*	*n.s.*	
Experiment 2	Control	27.5 a	13.3 b	72.5	1.8	12.4 a
	Calcium chloride	12.8 b	14.2 b	84.8	0	1 b
	Sodium silicate	15.7 b	30.0 a	67.2	0.9	1.9 b
	Significance	*	**	*n.s.*	*n.s.*	*
Experiment 4	Control	58.2 a	0.9	91.1	3.6	4.4
	Calcium chloride	63.8 a	0.7	98.5	0.1	0.7
	Sodium silicate	41.3 b	0.6	98.7	0.1	0.6
	Significance	**[2]	*n.s.*	*n.s.*	*n.s.*	*n.s.*

[1]Different letters represent significant differences by the ANOVA test. [2]Significant at *P < 0.05, **P < 0.01); *n.s.*: not significant

Table 3 Effects of treatments on fruit weight, firmness, and soluble solids concentration

		Treatment			
		Control	Calcium chloride	Sodium silicate	*Significance*
Experiment 1	Fruit weight (g fruit^{-1})	10.2 a[1]	9.3 ab	9.0 b	*[2]
	Fruit Firmness[2] (kg cm^{-2})	0.8	0.9	0.9	*n.s.*
Experiment 2	Fruit weight (g fruit^{-1})	8.8 a	7.5 b	8.6 a	*
	Soluble Solids (°Brix)	13.2 b	11.7 c	14.3 a	***
Experiment 3	Fruit weight (g fruit^{-1})	8.9	8.7	9.0	*n.s.*
	Soluble Solids (°Brix)	19.0 b	18.6 b	22.3 a	*
Experiment 4	Fruit weight (g fruit^{-1})	6.7	6.9	6.5	*n.s.*
	Fruit Firmness (kg cm^{-2})	0.5	0.57	0.6	*n.s.*
	Soluble Solids (°Brix)	16.1	15.4	16.3	*n.s*

[1]Different letters represent significant differences by the ANOVA test. [2]Significant at *P < 0.05, ***P < 0.001); *n.s.*: not significant

[2]Determined on the whole fruit using the penetrometer Effegi, FT 011, tip ø 5 mm

Table 4 Effects of treatments on the cherry skin color parameters measured on fruits of Experiment 1

Treatment	Color components		
	L[1]	S	H
Control	30.2 a[2]	19.6	6.3 a
Calcium chloride	30.2 a	20.9	6.6 a
Sodium silicate	29.4 b	15.7	4.0 b
Significance	**[3]	*n.s.*	*

[1]Lightness (L), saturation (S), and hue (H)

[2]Different letters represent significant differences by the ANOVA test. [3]Significant at *P < 0.05, **P < 0.01); *n.s.*: not significant

3.1.4 Experiment 4

Cracked fruits were higher than 50%, except in plants treated with the Si source (Table 2). Only the latter treatment reduced the percentage of damaged fruit compared to the control, whereas calcium chloride was not effective. On cracked fruits, most of the fractures were localized in the peduncular position, with no differences induced by treatments (Table 2). The analysis of the main quality parameters did not show differences due to treatments (Table 3).

3.2 Cryo-SEM and energy-dispersive X-ray microanalysis of fruits subjected to sodium silicate treatments

The analysis of frozen-hydrated samples of cherry fruits subjected to sodium silicate treatments by Cryo-SEM and energy-dispersive X-ray microanalysis demonstrated the presence of Si deposits on the surface of the fruits (Fig. 1A). The assay determined that even with the presence of sodium silicate on the outer surface of the fruit in the form of droplets remained following treatment (Fig. 1A and B), the cells

below the cuticle do not show a detectable amount of Si (Fig. 1C), whose silicon X-ray signal is always maintained below the level of the background (Fig. 1D, SiK, in green). This was observed by analyzing the cells of the mesocarp (Fig. 1C) and those of the epidermis, even at a distance of 50–60 µm from the drops of sodium silicate remaining on the surface (Fig. 1B). The same results were obtained from the analysis of frozen-hydrated samples of cherry fruits subjected to calcium chloride treatments (data not shown).

4 Discussion

Our results, obtained in different experimental conditions conducive to cracking, showed that canopy-applied sodium silicate during fruit ripening was effective in reducing sweet cherry cracking at harvest (Table 2). The effectiveness of sodium silicate was similar or higher than calcium chloride (Table 2), the most adopted technique for mitigating cracking.

In sweet cherry, three types of cracking have been described (Christensen 1996): in the cheek, in the apical end, and in the stem cavity region of the fruit. Multiple cracking, resulting from the combination of these three types, often occurs. Our data suggest that meteorological conditions influenced the incidence and type of cracking (Tables 1 and 2). Data indicate that high rainfall (Table 1) increased the occurrence of cracking in the stem cavity (Table 2). When low rainfall and low humidity occurred, no fruit cracking was observed. However, when rainfall was low but high humidity was recorded (Table 1), cracking was observed with the dominant type of cracking in the apical region (Table 2).

The results obtained via cryo-SEM and energy-dispersive X-ray microanalysis indicate that Si was localized only on the surface of the fruit, without any significant detection of its penetration into internal tissues (Fig. 1). However, some Si in traces, at concentrations below the resolution capability of the instrument, could be present in the observed tissues.

Although Si is not considered an essential element for plant nutrition and its questionable role in plant biology have been reported (Coskun et al. 2019), many authors have shown its beneficial effects in a variety of species and environmental conditions (Ma and Yamaji 2006; Coskun et al. 2019). The favorable effects of Si on the crop are often associated with the reinforcement of the cell walls due to the deposition of Si in the form of amorphous silica and opal phytoliths (Epstein 1999). Si displays a notable ability to increase the extensibility of the cell wall, a prerequisite for growth (Ma and Takahashi 1993). The application of Si to rice seedlings promoted plant growth mainly by enhancing cell elongation of the epidermal cells in the young shoot tissues and increased cell wall extensibility (Hossain et al. 2002).

When treated with Si, cell walls of sorghum roots were strengthened in the mature basal region, and cell walls in the apical and subapical zones showed an increase in extensibility, which resulted in a promotion of root elongation (Hattori et al. 2003). Si has also been reported to improve tissue extensibility and enhance cell enlargement in cucumber plants under salt and drought stress (Ouzounidou et al. 2016).

Si can modify the mechanical properties of tissues, contributing to cell wall rigidity and reinforcement, as it interacts with primary cell wall constituents (Currie and Perry 2007); at the same time, it increases the elasticity of the cell wall during distension and extension growth (Pilon-Smits et al. 2009).

Our data (Table 3; Fig. 1) indicate that canopy-applied Si did not build up a protective film covering the whole fruit surface, which would have prevented water exchanges between the fruit and the outside slowing down phloem translocation and consequent accumulation of sugars in the fruit. Such effects were not recorded in any of the experiments (Table 3).

The reduction of cracks also plays a role in the reduction of pathogen incidence (Børve et al. 2000). Therefore, Si applied to the canopy could also reduce the incidence of monilia (*Monilinia spp.*) and *Botrytis cinerea* on fruits. Si forms a physical barrier and stimulates defense reactions against pathogens (Ma and Takahashi 2002; Ma and Yamaji 2006; Wang et al. 2017; Bat-Erdene et al. 2021).

Additionally, Si treatment influenced color (Experiment 1; Table 4), reducing hue and brightness. Chromatic functions of chroma and hue are negatively correlated with anthocyanin levels in the fruit skin (Gonçalves et al. 2007), indicating higher concentrations of anthocyanins in the berry skin. In apples, canopy-applied Si has been shown to stimulate anthocyanin accumulation and induce skin color traits (Karagiannis et al. 2021).

Canopy-applied Si resulted in a significant increase in total soluble solids of cherry fruits (Experiments 2 and 3; Table 3), an effect attributable to the stimulating action of Si on the leaf photosynthetic activity (Ma and Takahashi 2002; Ma and Yamaji 2006; Bat-Erdene et al. 2021).

Research outcomes on fruit cracking highlight the relevance of mechanical properties of fruit epidermal cell wall strength, overall extensibility, elasticity, and cuticular membrane composition (Alkio et al. 2012; Balbontín et al. 2014; Quero-García et al. 2021). An elastic epidermis will increase the tolerance to cracking since it is more capable of accommodating the increase in flesh volume during fruit growth and reducing the formation of cuticular fractures.

Si appears to be a good candidate to achieve both of these conditions in the fruit. Therefore, these parameters should be included in further experiments.

5 Conclusions

Fruit cracking is a serious concern for cherry growers worldwide. Si is a beneficial element, known for its effectiveness against various biotic and abiotic stresses and therefore of interest in sustainable cherry cultivation. The current study highlights how canopy-applied Si sources may effectively contribute to reducing cherry cracking, acting as an alternative technique to other preventive methods. Future studies should gain further knowledge on Si mode of actions and Si nutrition in different cherry rootstocks and cultivars.

Acknowledgements Authors gratefully acknowledge Nino Quartieri farm (Savignano sul Panaro, MO), Neri-Vezzali farm (Vignola, MO), Testoni farm (Castelfranco Emilia, MO), and the Research Center of the Michigan State University (Michigan, USA) for hosting the field trials. The authors kindly acknowledge Prof. Dr. Jian Feng Ma and Dr. Alessandra Lombini for the critical reading of the manuscript.

Author Contribution ADR conceived the study, designed and performed the experiments, and wrote the manuscript. MQ contributed to performing the experiment and to the revision of the manuscript. ARD contributed to the writing of the manuscript. AM performed the Cryo-SEM and energy-dispersive X-ray analysis and interpretation and contributed to the writing. LS conceptualized the Cryo-SEM and energy-dispersive X-ray analysis and contributed to the revision of the manuscript. GS performed the experiments and contributed to the revision of the manuscript. All authors have read and approved the final manuscript.

Funding Open access funding provided by Alma Mater Studiorum - Università di Bologna within the CRUI-CARE Agreement.

Declarations

Conflict of Interest The authors declare no competing interests.

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