

Proceeding



Effect of calcium and seaweed based biostimulant on sweet cherry profitability and quality ⁺

Marlene Santos ^{1,*}, Carolina Maia ², Inês Meireles ², Sandra Pereira ¹, Marcos Egea-Cortines ³, João Ricardo Sousa ^{1,4}, Fernando Raimundo ^{1,4}, Manuela Matos ^{1,5} and Berta Gonçalves ^{1,4}

- ¹ Centre for Research and Technology of Agro-Environmental and Biological Sciences, CITAB, Inov4Agro, University of Trás-os-Montes and Alto Douro, UTAD, Quinta de Prados, 5000-801 Vila Real, Portugal. mpsantos@utad.pt (M.S.); sirp@utad.pt (S.P.); jricardo@utad.pt (J.R.S.); fraimund@utad.pt (F.R.); mmatos@utad.pt (M.M.); bertag@utad.pt (B.G.)
- ² University of Trás-os-Montes e Alto Douro, 5000-801 Vila Real, Portugal; carolina.maia98@gmail.com (C.M.); inesmeireles24@gmail.com (I.M.)
- ³ Instituto de Biotecnología Vegetal, Universidad Politécnica de Cartagena, 30202 Cartagena, Spain; marcos.egea@upct.es (M.E-C.)
- ⁴ Department of Biology and Environment (DeBA), University of Trás-os-Montes e Alto Douro, 5000-801 Vila Real, Portugal
- ⁵ Department of Genetics and Biotechnology (DGB), University of Trás-os-Montes e Alto Douro, 5000-801 Vila Real, Portugal
- * Correspondence: mpsantos@utad.pt
- + Presented at the 3rd International Electronic Conference on Agronomy, 15-30 October 2023.

Abstract: Sweet cherry tree is one of the most important crops worldwide, producing fruits with high economic importance due to its nutritional value and bioactive properties, with benefits to human health. Due to the currently unstable climatic conditions, cherry cracking has become a significant disorder, strongly affecting the quality and yield of cherry orchards. A cracking rate of 20-25% at harvest can render cherry production unprofitable, decreasing the commercial value of the fruit, as only the cracked ones can be sold to processing industries. This study aims to assess the impact of calcium and seaweed-based biostimulant applications on sweet cherry quality and profitability in cv. Sweetheart. Applying 300 g hL⁻¹ of calcium led to a significant 52% reduction in the cracking index and a substantial 136% increase in orchard yield. Similarly, applying 150 mL hL⁻¹ of seaweed resulted in a 2% increase in fruit weight and a 3% decrease in the cracking index. Therefore, our findings suggest that calcium and seaweed-based biostimulant could serve as novel and sustainable alternatives for orchard producers, enhancing cherry profitability and marketability.

Keywords: agricultural biofortification; calcium; cracking; crop nutrition; fruit quality; productivity; seaweed; sweet cherry

Published: date

name

staff during production.

Publisher's Note: MDPI stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Citation: To be added by editorial

Academic Editor: Firstname Last-



Copyright: © 2022 by the authors. Submitted for possible open access publication under the terms and conditions of the Creative Commons Attribution (CC BY) license (https://creativecommons.org/license s/by/4.0/).

1. Introduction

Sweet cherry (*Prunus avium* L.) is a fleshy fruit highly affected by cracking, a severe physiological disorder with strong implication in the quality and profitability of cherry orchards, decreasing its marketability [1]. Cracking is difficult to study, even under controlled conditions, since it occurs due to a combination of genetic and environmental conditions [1-3]. Several compounds have been applied in the orchards trying to increase cherry quality and yield and decrease cracking by crop nutrition [4]. Calcium is an important nutrient to improve fruit quality [5], and has been reported to play an important role in reducing cracking susceptibility [6,7]. Biostimulants are natural compounds obtained from fungi, bacteria or marine algae-based seaweed extracts, representing a new approach to reduce cracking [4]. These substances represent a sustainable

alternative to the use of conventional chemicals, being correlated with an improvement on tolerance against abiotic stresses and enhancement of plant growth, leading to an increase of quality and yield on agricultural crops [8,9]. Thus, this study intended to apply calcium and seaweed based biostimulant (*Ascophyllum nodosum*) at foliar level in sweet cherry trees cv. Sweetheart, trying to increase cherry quality and profitability by crop nutrition.

2. Material and Methods

2.1. Experimental design

This study was carried out during 2021 in an orchard located in Santa Eulália, São Martinho de Mouros, Resende (41°04'55.3"N 7°53'35.2"W, altitude 615 m). In order to analyze the cherry quality and profitability by crop nutrition and try to decrease the cherry cracking, the cultivar Sweetheart was selected to perform this trial, where calcium and seaweed based biostimulant (*Ascophyllum nodosum*) were applied at foliar level. The trees were spaced 4 m x 4 m (corresponding to 625 trees/ha), being selected 12 trees to apply each treatment, namely two concentrations of calcium (Kit Plant Ca), 300 g hL⁻¹ (Ca_300) and 150 g hL⁻¹ (Ca_150), two concentrations of seaweed based biostimulant (Foralg), 150 mL hL⁻¹ (Seaweed_150) and 75 mL hL⁻¹ (Seaweed_75), a combination of 300 g hL⁻¹ of calcium and 150 mL hL⁻¹ of seaweed or calcium. From the 12 trees of each treatment, fruits were collected at the commercial ripening stage.

2.2. Biometric parameters (fruit weight and larger diameter)

The biometric parameters were analyzed in 30 fruits randomly collected from each treatment, using an electronic balance (EW2200-2NM, Kern, Germany) to determine the fruit weight (g) and a digital caliper (Mitutoyo, Hampshire, UK) to determine the fruit size (mm), specifically the larger diameter.

2.3. Cracking index

The induced cracking index (CI) was determined as described by Christensen, 1972 [10]. For this, 3 replicates of 50 fruits without cracking from each treatment were immersed in 2 L of distilled water. After 2, 4, and 6 h, the fruits were observed to check the presence of macroscopic cracks. In each observation, the cracked fruits were removed while the fruits without cracks were kept in the water. At the end, considering the number of cracked cherries after 2, 4, and 6 h of immersion in water (corresponding to a, b, and c, respectively), the CI was determined as:

CI = ((5a + 3b + c) * 100)/250

2.4. Orchard yield

At the commercial ripening stage, the production per tree was determined (kg/tree), including the amount of healthy and unhealthy cherries. Using the production of the 12 trees per treatment, the total production (kg) as well as the percentage of healthy and unhealthy cherries within each treatment was also evaluated. Lastly, based on total production per tree and number of trees per ha, the productivity (t/ha) was estimated as:

Productivity = ((total production/tree) * (number of trees/ha) / 1000)

2.5. Statistical analysis

The statistical analysis using one-way analysis of variance (ANOVA), followed by Tukey's post hoc multiple range test (p < 0.05) was carried out in the Software SPSS V.27 (SPSS-IBM, Corp., Armonk, New York, USA).

3. Results and Discussion

3.1. Biometric parameters (fruit weight and larger diameter)

By the evaluation of fruit size parameters, it was observed a slightly increase on fruit weight and larger diameter in cherries treated with 150 mL hL⁻¹ of seaweed, while cherries treated with 300 g hL⁻¹ of calcium presented the lower fruit weight and larger diameter (data not shown). The analysis of both parameters revealed significant differences among the different treatments (p < 0.001). Comparing the fruit size with the control (Figure 1), cherries treated with 150 mL hL-1 of seaweed represents the unique treatment that leaded to an increase of fruit size, with an increment of 1.90% in fruit weight and 0.64% in fruit larger diameter. In contrast, the other treatments caused a decrease in fruit size. Cherries treated with 300 g hL-1 of calcium presented the higher reduction in fruit size, followed by the combination of both nutrients, treatment with 150 g hL⁻¹ of calcium and cherries treated with 75 mL hL⁻¹ of seaweed. Thus, fruit weight showed a decrease of 11.15% in Ca 300, 9.49% when both nutrients were combined, 6.25% in Ca_150, and 1.82% in Seaweed_75. Concerning to the fruit larger diameter, it was observed a decrease of 4.23% in Ca_300, 2.84% in combination of both nutrients, 2.10% in Ca_150, and 1.22% in Seaweed_75. Similar results were obtained by Correia et al. (2015), whose seaweed application in cvs. Sweetheart and Skeena results in an increase of fruit dimensions, both in weight and diameter [11]. The application of Ascophylum nodosum in cv. Staccato also resulted in bigger fruits and with similar weight and diameter compared to control cherries [12]. In cvs. Kordia and Regina, the application of a plant extract biostimulant also increased the fruit diameter [13].



Figure 1. Fruit weight (A) and fruit larger diameter (B) relatively to control treatment. In each treatment, the value corresponds to a percentage (%) of increase or decrease of fruit growth comparatively to the control.

3.2. Cracking Index

The analysis of CI data showed significant differences among the different treatments (p < 0.001), in which cherries treated with 300 g hL⁻¹ Ca presented the lowest CI, while the combination of both nutrients presented the highest CI (data not shown). Thus, comparing the CI with the control (Figure 2), it was observed an increase of 27.47% when both nutrients were applied and a decrease of 3.14%, 14.86%, 44.59%, and 51.79% for Seaweed_150, Seaweed_75, Ca_150, and Ca_300, respectively. Previous work reported a decrease around 50% in CI, when calcium was applied in cv. Sweetheart [6]. Calcium treatments also reduced the CI in cv. Ferrovia [7]. Likewise, the application of *Ascophyllum nodosum* also decreased CI in cvs. Sweetheart and Skeena [11].





3.3. Orchard yield

3.3.1. Production per tree

The total production per tree as well as the amount of healthy and unhealthy cherries per tree (Figure 3) were higher in cherries treated 300 g hL⁻¹ of calcium (42.98±18.24, 36.47±16.06, and 6.51±2.87 kg/tree, respectively) and lower in cherries treated with 150 mL hL⁻¹ of seaweed (9.17±4.87, 7.47±4.10 and 1.70±0.94 kg/tree, respectively). The analysis of these parameters revealed significant differences among treatments (p < 0.001).



Figure 3. Total cherry production per tree (A), amount of healthy cherries per tree (B) and amount of unhealthy cherries per tree (C) in each treatment. Each column is expressed as mean \pm SE (n = 12). Different letters mean significant statistical differences (p < 0.001) according to Tukey's test.

3.3.2. Total production

Considering the production of the 12 trees of each treatment, the total production was determined as well as the amount of healthy and unhealthy cherries. Cherries treated with 300 g hL⁻¹ of calcium had a higher total production (515.71 kg), corresponding to 437.64 kg of healthy cherries and 78.06 kg of unhealthy cherries (Figure 4). In contrast, the foliar application of 150 mL hL⁻¹ of seaweed resulted in the lowest total production (110.00 kg), corresponding to 89.64 kg and 20.35 kg of healthy and unhealthy cherries,

respectively. Additionally, the amount of healthy cherries in Ca_300 corresponded to 84.86% of the total production, while in Seaweed_150 treatment the amount of healthy cherries corresponded to 81.50% of the total production (Figure 5).



Figure 4. Total production, amount of healthy and unhealthy cherries (kg) obtained from the 12 trees of each treatment.



Figure 5. Percentage (%) of healthy and unhealthy cherries obtained from the 12 trees of each treatment.

Comparing the total production in each treatment with the control (Figure 6), the Ca_300 treatment presented the highest increase of total production (135.80%), followed by Ca_150 treatment (55.91%) and the combination of both nutrients (37.90%). On the other side, treatments with seaweed leaded to a decrease in total production (49.71% for Seaweed_150 and 10.80% for Seaweed_75).



Figure 6. Total production relatively to control. In each treatment, the value corresponds to a percentage (%) of increase or decrease of total production comparatively to the control. .

3.3.3. Productivity (t/ha)

Significant differences among the different treatments (p < 0.001) were found in productivity (Figure 7). Higher yield was found in cherries treated with 300 g hL⁻¹ Ca (26.86±11.40 t/ha), followed by treatment with 150 g hL⁻¹ Ca (17.76±7.82 t/ha), treatment that combines both nutrients (15.71±7.72 t/ha) and control (11.39±7.26 t/ha). The application of seaweed resulted in lower productivity, 10.16±9.02 t/ha for seaweed at 75 mL hL⁻¹ and 5.73±3.04 t/ha for seaweed at 150 mL hL⁻¹.



Figure 7. Cherry productivity (t/ha) in each treatment. Each column is expressed as mean \pm SE. Different letters mean significant statistical differences (*p*<0.001) according to Tukey's test.

The use of biostimulants have been associated with an increase of plant growth and yield in several crops [14]. As reported by Correia et al. (2020), the application of *Ascophyllum nodosum* allowed to increase the yield of sweet cherry trees of cv. Skeena [15]. In cvs. Kordia and Regina, the foliar application of a plant extract biostimulant also leaded to an increase of the fruit yield [13]. In our study, the application of a seaweed based biostimulant resulted in lower fruit yield, contrasting with the results described in the literature. However, in this study we used another cultivar, Sweetheart, that can be less responsive to the treatments with seaweed. On the other hand, calcium has been reported as an essential nutrient in several plants, especially in fruits [7]. Our results showed a yield increasement when the seaweed was combined with calcium, and also when calcium was applied individually, which was also attested by Correia et al. (2020) in cv. Skeena [15].

4. Conclusions

Consumers prefer fruits with good size and without defects, whereby fruit size and cracking index are important quality parameters with high impact on fruits marketability. In our study, the pre-harvest application of calcium and seaweed based biostimulant has proven to have a positive effect on cherry quality and yield. Both nutrients played a significant role in decreasing cracking index, especially in cherries treated with calcium. On the one hand, the use of *Ascophyllum nodosum* produced bigger fruits, but with lower orchard yield. On the other hand, calcium application generated smaller fruits, but highly increased the orchard yield in addition to a significative decrease on cracking index. Despite new studies and strategies are needed, these findings suggest that the nutrients under study could represent new and sustainable alternatives to be used by producers in their orchards to improve cherry profitability and marketability.

Author Contributions: Conceptualization, M.S. and B.G.; Methodology and field work, M.S., C.M., I.M., and S.P.; Compounds application in the field, J.R.S. and F.R.; Data analysis, M.S.; Writing—original draft preparation, M.S.; Writing—review and editing, C.M., I.M., S.P., M.E-C., J.R.S., F.R., M.M., and B.G.; Supervision, M.E-C., M.M., and B.G.; Funding acquisition, B.G. All authors have read and agreed to the published version of the manuscript.

Funding: This research was supported by the European Agricultural Fund or Rural Development (EAFRD) and by the Portuguese State in the context of action 1.1. Grupos Operacionais integrado

na medida 1. Inovação do PDR 2020–Programa de Desenvolvimento Rural do Continente–Grupo Operacional para a valorização da produção da Cereja de Resende e posicionamento da subfileira nos mercados (iniciativa nº 362).

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

Acknowledgments: Marlene Santos acknowledges the financial support provided by FCT - Portuguese Foundation for Science and Technology (PD/BD/ 150257/2019), under the Doctoral Program 'Agricultural Production Chains – from fork to farm' (PD/00122/2012). The authors also acknowledge the support by National Funds from FCT - Portuguese Foundation for Science and Technology, under the project UIDB/04033/2020 (CITAB research unit).

Conflicts of Interest: The authors declare no conflict of interest.

References

- 1. Santos: M.; Egea-Cortines, M.; Gonçalves, B. and Matos, M. Molecular mechanisms involved in fruit cracking: A review. Frontiers in Plant Science 2023, 14, 1130857. DOI: 10.3389/fpls.2023.1130857.
- Khadivi-Khub, A. Physiological and genetic factors influencing fruit cracking. Acta Physiologiae Plantarum 2015, 37, 1718. DOI: 10.1007/s11738-014-1718-2.
- Rehman, M.U.; Rather, G.H.; Dar, N.A.; Mir, M.M.; Iqbal, U.; Mir, M.R.; Fayaz, S. and Hakeem, K.R. Causes and prevention of cherry cracking: A review. In: Crop Production and Global Environmental Issues. Hakeem KR, editor. Cham: Springer International Publishing; 2015. p. 543-552. DOI: 10.1007/978-3-319-23162-4_19.
- 4. Correia, S.; Schouten, R.; Silva, A.P. and Gonçalves, B. Sweet cherry fruit cracking mechanisms and prevention strategies: A review. Scientia Horticulturae 2018, 240, 369-377. DOI: https://doi.org/10.1016/j.scienta.2018.06.042.
- 5. Winkler, A. and Knoche, M. Calcium and the physiology of sweet cherries: A review. Scientia Horticulturae 2019, 245, 107-115. DOI: https://doi.org/10.1016/j.scienta.2018.10.012.
- Correia, S.; Santos, M.; Glińska, S.; Gapińska, M.; Matos, M.; Carnide, V.; Schouten, R.; Silva, A.P. and Gonçalves, B. Effects of exogenous compound sprays on cherry cracking: skin properties and gene expression. Journal of the Science of Food and Agriculture 2020, 100, 2911-2921. DOI: https://doi.org/10.1002/jsfa.10318.
- Michailidis, M.; Polychroniadou, C.; Kosmidou, M.-A.; Petraki-Katsoulaki, D.; Karagiannis, E.; Molassiotis, A. and Tanou, G. An early calcium loading during cherry tree dormancy improves fruit quality features at harvest. Horticulturae 2021, 7, 135. DOI: https://doi.org/10.3390/horticulturae7060135.
- 8. Bulgari, R.; Franzoni, G. and Ferrante, A. Biostimulants application in horticultural crops under abiotic stress conditions. Agronomy 2019, 9, 306. DOI: https://doi.org/10.3390/agronomy9060306.
- 9. Drobek, M.; Frąc, M. and Cybulska, J. Plant biostimulants: importance of the quality and yield of horticultural crops and the plant improvement of tolerance to abiotic stress-Areview. Agronomy 2019. 9. 335. DOI: https://doi.org/10.3390/agronomy9060335.
- 10. Christensen, J.V. Cracking in cherries. Acta Agriculturae Scandinavica 1972, 22, 128-136. DOI: 10.1080/00015127209433471.
- Correia, S.; Oliveira, I.; Queirós, F.; Ribeiro, C.; Ferreira, L.; Luzio, A.; Silva, A.P. and Gonçalves, B. Preharvest Application of Seaweed Based Biostimulant Reduced Cherry (Prunus Avium L.) Cracking. Procedia Environmental Sciences 2015, 29, 251-252. DOI: https://doi.org/10.1016/j.proenv.2015.07.187.
- 12. Gonçalves, B.; Morais, M.C.; Sequeira, A.; Ribeiro, C.; Guedes, F.; Silva, A.P. and Aires, A. Quality preservation of sweet cherry cv. 'staccato' by using glycine-betaine or Ascophyllum nodosum. Food Chemistry 2020, 322, 126713. DOI: https://doi.org/10.1016/j.foodchem.2020.126713.
- 13. Basile, B.; Brown, N.; Valdes, J.M.; Cardarelli, M.; Scognamiglio, P.; Mataffo, A.; Rouphael, Y.; Bonini, P. and Colla, G. Plantbased biostimulant as sustainable alternative to synthetic growth regulators in two sweet cherry cultivars. Plants (Basel, Switzerland) 2021, 10, 619. DOI: 10.3390/plants10040619.
- 14. Ali, O.; Ramsubhag, A. and Jayaraman, J. Biostimulant properties of seaweed extracts in plants: implications towards sustainable crop production. Plants 2021, 10, 531. DOI: https://doi.org/10.3390/plants10030531.
- Correia, S.; Queirós, F.; Ferreira, H.; Morais, M.C.; Afonso, S.; Silva, A.P. and Gonçalves, B. Foliar application of calcium and growth regulators modulate sweet cherry (Prunus avium L.) tree performance. Plants 2020, 9, 410. DOI: 10.3390/plants9040410.