Effect of calcium nitrate treatment on reduction of berry shattering in ‘Cheongsoo’ grape cultivars

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Abstract

This study was conducted to examine the effect of calcium nitrate (CaNO$_3$) on berry shattering in and the fruit characteristics of the ‘Cheongsoo’ grape cultivar. The rate of berry shattering was reduced regardless of CaNO$_3$ concentration when the detachment force of the abscission zone at the stalk-berry junction increased. When 0.8% CaNO$_3$ was treated, the berry shattering rate decreased by approximately 40% compared to the control; this was the lowest value. Furthermore, fruit quality declined and side effects were not observed following the CaNO$_3$ treatment although fruit firmness also increased by 57% compared to the control. These results indicate that treatment with CaNO$_3$ can contribute to stable production of ‘Cheongsoo’ grapes.

Keywords: abscission; ‘Cheongsoo grape’; detachment force; fruit characteristics; tensile strength

Introduction

Grapes are regarded as the fourth most important fruit crop in Korea. In 2020, the cultivated area of grapes in Korea was 12,676 ha, with the ‘Campbell Early’, ‘Kyoho’, and ‘Shine Muscat’ varieties accounting for more than 90% of this (Kim et al., 2021). This can be problematic because it may be difficult to meet the demands of consumers interested in diversity while still maintaining competitive pricing. Therefore, it is necessary to introduce cultivars that can be used for fresh consumption and to establish a sustainable grape industry and revitalize the grape market (Heo et al., 2017).

In Korea, many efforts have been made to breed and use new grape cultivars (Park et al., 2017; Kwon et al., 2019; Park et al., 2020; Kim et al., 2020). Researchers of Rural Development Administration in Korea have developed a promising cultivar ‘Cheongsoo’ by crossing ‘Siebel 9110’ and ‘Himrod’ varieties. It has excellent fruit quality and flavor, is suitable for fresh consumption, and has optimal conditions for wine production (Chang et al., 2014). For these reasons, its cultivation area has been expanding, yet vineyards have reported reduced yield due to berry shattering. It is important to develop a practical method to reduce berry shattering to promote stable grape production of ‘Cheongsoo’.

Berry shattering occurs in grapes for abscission-related factors and is known to be strongly related to detachment force of the abscission zone at the stalk-berry junction (Li et al., 2020). The detachment force can...
be increased with calcium nitrate (Ca(NO$_3$)$_3$); however, the effect of Ca(NO$_3$)$_3$ on berry shattering and fruit characteristics differs by concentration. Hence, we tested whether foliar fertilization with Ca(NO$_3$)$_3$ could be used as a method for minimizing berry shattering in ‘Cheongsoo’ and attempted to select an optimal treatment concentration of the same.

**Materials and Methods**

**Background information**

This study was performed using 5-year-old ‘Cheongsoo’ grapes planted at 3 m in row and 3 m in vines at the vineyard of Gangwondo Agricultural Research and Extension Services located in Chuncheon, Korea. For this experiment, Ca(NO$_3$)$_3$ concentrations were set to 0.0, 0.2, 0.4, and 0.8%. At the veraison period, each concentration of Ca(NO$_3$)$_3$ was sprayed once onto leaves and clusters with 70 to 75 berries whose fruiting was controlled.

**Quantitative analysis**

The effect of Ca(NO$_3$)$_3$ treatment on the reduction of berry shattering was examined by testing the berry shattering ratio and detachment force of clusters at optimal harvest period. The berry shattering ratio of berries was calculated as (the number of falling berries / the total number of berries) × 100. Detachment force can be evaluated by the tensile strength of the abscission zone at the stalk-berry junction, and was measured using a rheometer (Sun Scientific, Tokyo, Japan). Fruit characteristics after administration of Ca(NO$_3$)$_3$ were observed by evaluating berry weight, total soluble solids (TSS), titratable acidity (TA), and fruit firmness as these are regarded as the main indicators of fruit characteristics in grapes (Heo and Park, 2016). For measurement of cluster weight, five clusters from three vines were randomly harvested and each cluster was weighed using a digital scale. Then, berry weight was calculated by dividing cluster weight by the number of berries per cluster. TSS was measured with a digital refractometer after extracting fruit juice from ten berries from each cluster (expressed in °Bx). TA was measured for sugar content using an automatic titrator (Schott, Mainz, Germany) after adding 5 ml of the fruit juice obtained to 15 ml of distilled water (Heo et al., 2015). Titration was conducted with 0.1 N NaOH to a pH of 8.2 and was expressed as a value converted into tartaric acid. For measurement of fruit firmness, five berries were randomly selected from each cluster and the rind from the equator was removed. Then, the maximum pressure was measured using a texture analyzer equipped with a 2 mm diameter plunger, set to a speed of 2 mm·s$^{-1}$ and a depth of 5 mm (expressed in Newton (N)).

**Statistical analysis**

Correlation analysis was performed to test the relationship between berry shattering and detachment force in the ‘Cheongsoo’ cultivar. Statistical comparison of berry shattering and fruit characteristics based on Ca(NO$_3$)$_3$ treatment concentration was performed using the analysis of variance and Duncan’s multiple range tests in the SPSS program (Version 28, IBM, USA). Correlation was deemed significant at $p = 0.01$.

**Results and Discussion**

Table 1 shows the effects of different concentrations of Ca(NO$_3$)$_3$ on the berry shattering and fruit characteristics of the ‘Cheongsoo’ cultivar. The berry shattering rate decreased regardless of treatment concentration when Ca(NO$_3$)$_3$ was administered. When treated with Ca(NO$_3$)$_3$ at 0.4% and 0.8%, the detachment force increased sharply to 47.2% and 44.9%, respectively, compared to the control. In contrast, the berry shattering rate decreased considerably. Among the important indicators, the berry shattering rate was only correlated with detachment force (Table 2).
Table 1. Effect of CaNO3 treatment on berry shattering and fruit characteristics in 'Cheongsoo' grapes

| Concentration (%) | Berry shattering rate (%) | Detachment force (g/cm²) | Berry weight (g) | Total soluble solids (°Brix) | Titratable acidity (%) | Fruit firmness (N) |
|-------------------|--------------------------|--------------------------|-----------------|-----------------------------|------------------------|--------------------|
| 0.0               | 23.0 a                   | 116.5 b                  | 2.7 a           | 17.3 c                     | 0.88 a                 | 0.14 c             |
| 0.2               | 19.4 b                   | 120.7 b                  | 2.6 a           | 18.7 a                     | 0.59 b                 | 0.18 b             |
| 0.4               | 16.3 bc                  | 171.6 a                  | 2.6 a           | 18.2 b                     | 0.58 b                 | 0.22 a             |
| 0.8               | 13.9 c                   | 168.9 a                  | 2.7 a           | 17.6 c                     | 0.85 a                 | 0.22 a             |

The same letters denote no significant difference at p < 0.05.

Table 2. Correlation matrix for study variables

| Pearson correlation | Berry shattering rate | Detachment force | Total soluble solids | Titratable acidity | Fruit firmness |
|---------------------|-----------------------|------------------|----------------------|-------------------|----------------|
| Berry shattering rate | 1                    |                  |                      |                   |                |
| Detachment force | -0.382**              |                  |                      |                   |                |
| Total soluble solids | -0.086               |                  |                      |                   |                |
| Titratable acidity | -0.004                |                  | -0.431**             |                   |                |
| Fruit firmness | -0.160                | 0.492**           | 0.149                | -0.159            | 1              |

** Correlation is significant at the 0.01 level.

The relationship between berry shattering and detachment force reported by Wu et al. (2008) and Sabir and Sabir (2017) suggested that one of the main causes of the former in the ‘Cheongsoo’ cultivar is decreased detachment force of the abscission zone at the stalk-berry junction. This is suspected to be caused by cell dissolution, separation, and breakdown in the abscission zone. Calcium in the cell wall ionically binds pectin in the middle layer of the cell. This increases the binding force of the middle layer of the cell, protecting the structure and function of the cell membrane and delaying cell breakdown caused by aging (Gao et al., 2019).

For this reason, calcium has been successfully used to mitigate berry shattering (Bassiony et al., 2018; Nireshkumar et al., 2020), berry cracking (Yu et al., 2020), and fruit softening (Gao et al., 2020). From this experiment, we also verified that CaNO3 would result reduce berry shattering given that the ‘Cheongsoo’ grapes contain calcium. However, it should be noted that treatments with 0.4% and 0.8% CaNO3 showed similar detachment force levels but differing berry shattering levels, with the former for the 0.8% CaNO3 treatment being more reduced. This result indicates that other factors may be associated with berry shattering. For example, ethylene production has been reported as another cause of berry shattering (Ye et al., 2017), and investigating it further would be helpful to more effectively prevent the problem in ‘Cheongsoo’ grapes.

Although CaNO3 treatment results in a reduction in berry shattering, it cannot be used in farms if it decreases fruit characteristics; therefore, it is also important to evaluate the latter. From this study, we confirmed that berry weight was not affected by CaNO3 treatment. In two treatment conditions (0.2% and 0.4% conc.), TSS increased considerably, with the highest value being 18.7 °Bx after 0.2% CaNO3 treatment. In addition, TA decreased significantly at concentrations of 0.2% and 0.4%. Similarly, the administration of CaNO3 improved TSS and decreased TA in "ruby seedless" grapes (Huang et al., 2018), suggesting that CaNO3 improves the taste of some grape cultivars. One study shows that shoot growth was suppressed and physiological activity increased in peach leaves treated with calcium, and that this improved fruit quality (Park et al., 2010). Similarly, TSS increases with CaNO3 treatment in ‘Cheongsoo’ grapes. The nutrients stored in the grape may become activated and more concentrated because CaNO3 might improve photosynthetic ability. At 0.8% CaNO3, TSS and TA remained at similar levels to those of the control. Although photosynthetic efficiency may increase with 0.8% CaNO3 treatment, the increased nitric acid might promote vegetative growth and thus negatively affect the balance of TSS and TA compared to 0.2% and 0.4% concentrations.
The results of this study show that berry firmness increases after CaNO₃ application. Many other studies have reported a significant relationship between calcium levels and fruit firmness in apples, blueberries, and peaches (Mohelpi et al., 2020; Shahid et al., 2020; Lobos et al., 2021). According to a recent study in grapes (Martins et al., 2020), the internal structure of the berry changes due to the expression of genes that prevent pectin degradation and cell wall softening, thus leading to berry firmness. From this study, CaNO₃ seems to result in a similar effect in 'Cheongsoo' grapes. It has also been reported that some fruit crops lose their commercial value owing to fruit cracking or rotting when calcium is foliar fertilized, but no side effect was observed when CaNO₃ was applied to 'Cheongsoo'. With treatment of 0.8% CaNO₃, berry shattering was reduced to a great extent while fruit quality was not reduced, thereby indicating that this treatment could be used effectively to solve the problem in 'Cheongsoo' grapes. It is expected that the foliar application of CaNO₃ fertilizer could be used to strengthen 'Cheongsoo' grapes that suffer from berry shattering.

**Conclusions**

In this study, the application of CaNO₃ not only reduced berry shattering in 'Cheongsoo', but also made a difference in fruit characteristics. It was determined that the effect varies according to treatment concentration. A higher concentration of CaNO₃ (0.8%) resulted in the lowest berry shattering rate yet did not decrease fruit quality. Medium concentrations of CaNO₃ (0.2% and 0.4%) decreased berry shattering rate slightly but greatly decreased titratable acidity. It would be useful to investigate the cause of these differences through additional studies to better understand CaNO₃ for use in 'Cheongsoo' grapes and other cultivars. Molecular studies using conditions that induce phenotypic differences are expected to play a major role in elucidating the genetic factors that control fruit quality and berry shattering. This will be important in the future, especially for breeding and cultivation of grapes.

**Authors' Contributions**

YSP: Conceptualization; Data curation; Investigation; Methodology; Project administration; Resources; Software; Writing - original draft. JCL, JHK and HNJ: Data curation; Formal analysis; Methodology; Validation. JYH: Formal analysis; Methodology; Supervision; Writing - review and editing. All authors read and approved the final manuscript.

**Ethical approval** (for researches involving animals or humans)

Not applicable.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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