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Reduction of postharvest losses of grapes depends on prevention of cracking during development and proper disinfection after harvest

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Table grapes are highly susceptible to postharvest losses due to infection by the gray mold fungus *Botrytis cinerea*. Protection of table grapes against decay during storage relies on a 'multiple hurdle approach', which entails using a combination of methods to reduce decay at critical control points. *Botrytis*, is a typical wound pathogen and the presence of cracks in the peel will enhance its potential to cause decay of table grapes. An important pre-harvest strategy to maintain table grapes during storage is to reduce the level of peel cracking before harvest. By applying the growth regulator gibberellic acid at late stages of fruit set, a significant reduction of cracking from over 40% to less than 10% was achieved. Consequently, postharvest losses were significantly reduced. Another major strategy to reduce losses after storage is to remove and disinfect the pathogens that may be present on the peel after harvest. The current commercial practice is to use SO_2 sheets which release the gas upon absorption of water during storage. However, until sufficient levels of SO_2 are established, the pathogen can enter the berries and thereby avoid exposure to the gas. We showed that application of ethanol solution after harvest reduced the level of postharvest decay. These reduced levels were comparable to application of SO_2 during storage. Our results demonstrate the importance in understanding the mode of infection, and by using a whole system approach, a significant impact in prevention of postharvest losses in table grapes can be attained.

Introduction

Grapes as a fleshy soft fruit have an inherent vulnerability to cracking. As in tomato, cracking in grapes can have a genetic propensity and physiological or horticultural triggers can induce interruptions in the peel. These form direct entry pathways for pathogens[3]. Cracks can either be long and deep and then they are termed 'macro-cracks', or small and shallow and then they are termed 'micro-cracks'. The microcracks can either transverse the cuticle, or remain superficial at the cuticle level. When cracks occur, there is a high risk of contamination by microorganisms residing on the surface. Given the presence of pathogens and the right environmental conditions the pathogens will start developing thus causing decay in the vineyard. If however, the fruit is protected by fungicides, or cracks have enough time to cure, the fruit may be saved. But there are situations where pathogens will penetrate or remain on the cracks and their growth will be arrested by natural antifungal compounds. The development of the pathogens will resume during storage when natural

defenses of the fruit gradually decline, resulting in significant postharvest losses. There are multiple ways in which we can intervene in the process one of which is to reduce the level of cracking using growth regulators that can modulate the structure of the peel during the early stages of fruit development. Gibberellic acid (GA) is widely used in seedless table grapes to complement for the lack of GA produced by the seeds and to enlarge the berries. The berry size of seeded table grape varieties is not affected by external GA[1], making them a good model to study the effect of GA on the peel. 'Zainy' is a local late ripening table grape variety with large berries, loose clusters, and high yield but with a propensity to cracking.

Another method to reduce the damage of cracking and the negative effects of opportunistic pathogens, is to disinfect the fruit after harvest. The postharvest disinfection approach by ethanol dips was developed by us together with American colleagues more than 10 years ago[2] but it has not been used commercially to-date partly because producers favor dry postharvest treatments rather than dipping, which requires an additional treatment of drying.

The objective of this paper is to bring to the attention of the audience of Volcani Voice the significance of reducing cracking in fruit and the importance of proper disinfection of the fruit surface prior to storage. These principles are true for many fruit types but the details are likely to change from system to system.

Results and Discussion

Morphological aspects of cracking

In sensitive grape varieties, cracks can often be observed on the peel (skin, exocarp) after veraison – the time point when the berries start to change color or soften and accumulate sugars. Macro-cracks are elongated cracks that can be readily observed without magnification (Fig. 1A). They can be linear and appear along the cheek, circular around the pedicel, or they can cross the bottom end of the berry. Opportunistic pathogens of the genus Alternaria or Cladosporium often use macro-cracks to initiate an infection in the vineyard (Fig. 1B) while *Botrytis cinerea* will often be the dominant pathogen after cold storage. Micro-cracks are expressed in several ways but often they are accompanied by browning due to oxidation of the injured tissue. In some cases, the cuticle along the crack can be sunken (Fig. 1C), likely because of dehydration. The same image can be observed by projecting light on it and observing the emitted light (auto-fluorescence). In the case of Fig. 1D the red background is the auto-fluorescence of the chlorophyll in the peel of the green berry and the cracks are decorated by green lining of phenolic compounds which accumulate around the crack. Clusters affected by cracking often develop browning and decay after cold storage (Fig. 1E).

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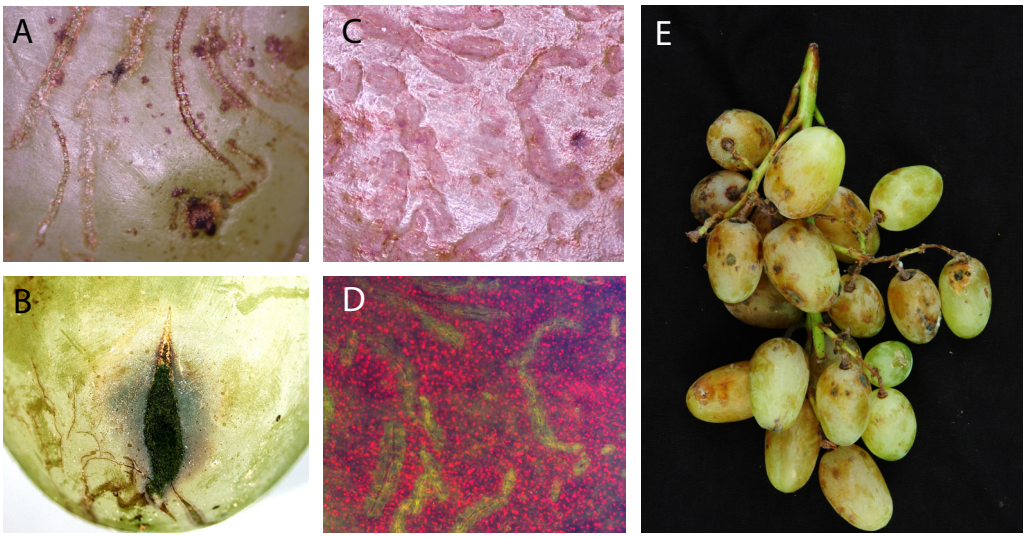

***Figure 1**: Macro and micro-cracks in 'Zainy' table grapes. A. Macro-cracks. B. Development of decay in the vineyard on a deep macro-crack. C. Surface of a berry with micro-cracks and sunken tissue along the crack. D. The same image as in C viewed by auto-fluorescence with excitation at 470 nm and emission above 500 nm. E. The appearance of 'Zainy' after storage without protection against decay.*

Reducing cracking with gibberellic acid and the consequences for decay after storage

In our studies we found that reproducible and statistically significant results for the reduction of cracking in 'Zainy' grapes were obtained through the application of gibberellic acid (Fig. 2A). GA was applied either at early fruit development stage, when berries were at diameter of 6 mm or at berry size of 8 to 10 mm. It should be emphasized that application of GA at 6 mm is the preferable stage for berry enlargement in seedless varieties. The control, untreated grapes, showed about 50% cracking, which is considered high but according to our experience with 'Zainy' not exceptional. While early treatment with GA had no significant effect, late treatment reduced decay to just above 10%, which translates to about a 4-fold reduction in cracking (Fig.2A). Dual treatment at both the early and late stages did not reduce cracking further, suggesting that the application stage is important. Application of the cytokinin, CPPU[4], did not reduce cracking significantly. We found that calcium, which is thought to reduce cracking, had no significant effect on berry cracking.

When cracks occur, there is a high risk of contamination by microorganisms residing on the surface

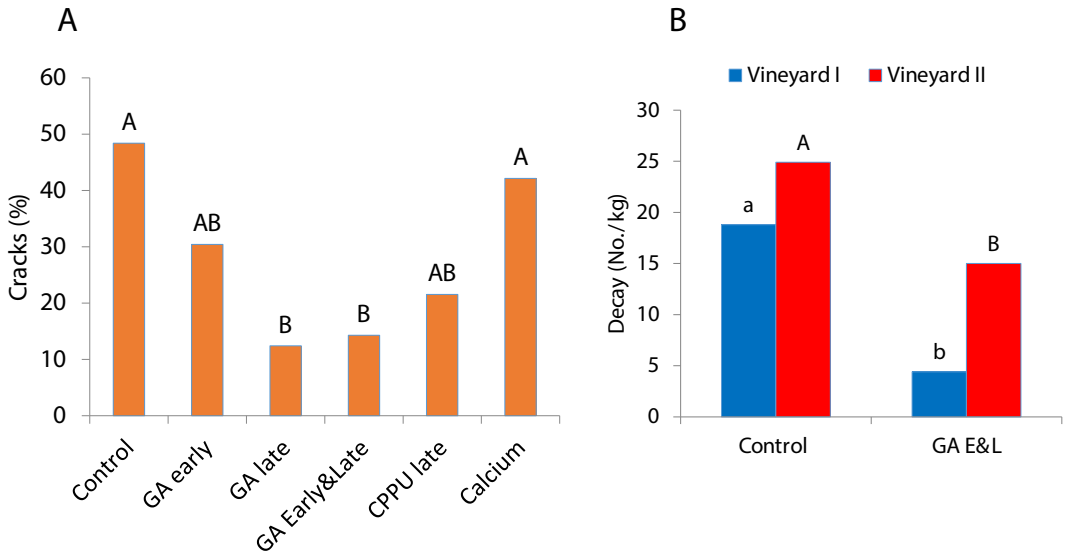

Figure 2: *Reduction of cracking and postharvest decay by gibberellic acid (GA).*
A. Application of GA (20 ppm with 0.025% triton-X100) at berry diameter of 6 mm (early), or 10 mm (late), or at both stages. The cytokinin CPPU (2 ppm with triton) was applied at 10 mm berry diameter. Calcium was applied 3 times at a rate of 0.4% as calnit.
B. Dual GA treatment at early and late fruit set (E&L) and control untreated fruit were stored at 0°C for 5 weeks and 3 d at 20°C. The experiment included fruit from two vineyards in Moshav Lachish and statistical analysis was performed separately for each vineyard shown as lowercase and uppercase letters above the bars.

In a subsequent experiment two vineyards were treated with a dual spray of GA aimed at ensuring treatment at a responsive stage of development. The first application was at berry diameter of 6 mm and the second, one week later. . The grapes from the two vineyards were stored at zero °C for 5 weeks after which they were evaluated for decay. The results from both vineyards demonstrate significant reduction in decay (Fig. 2B), confirming that reduction in cracking also improves the natural resistance of the berries to pathogens. It should be stated however that treatment with GA should not be considered as sufficient for commercial postharvest treatment against decay, as tolerance to decay in commercial practice is well below 1% and the treatment may only improve the efficacy of additional postharvest treatments.

Disinfection after harvest

If opportunistic pathogens encounter a natural opening such as a fresh crack, they will initiate filamentous growth into the berry. The commercial practice is to store the grapes with SO_2 paper sheets which protect against decay-development during storage. However, there is a window of time, from harvest until the initiation of SO_2 activity when the berries are less protected during which the disinfection technologies may play a significant role.

Clusters of 'Scarlotta' grapes were dipped in ethanol solution after harvest or stored in the presence of SO_2 sheets. This ethanol dipping treatment has a double action as it cleans the clusters and disinfects them. After 6 weeks of cold storage and 3 days at 20°C the clusters of the control treatment suffered high levels of decay compared to the treated clusters (Fig. 3). The number of decayed berries in the control was 60 which approximated 60% decay by weight. The SO_2 treatment, which reached a level of 6 to 8 ppm during storage, did not supply sufficient protection against decay with 10% of the berries suffering decay (severity was however much lower than the control). It should be stated that in commercial practice, the SO_2 treatment is considered very effective, with less than 1% decay after storage[2]. The ethanol dipping treatment gave in this case 3% decay with much cleaner appearance of the clusters. It should be emphasized that if *Botrytis* spores inoculated the flowers, no external disinfection treatment after harvest can prevent fruit decay or limit the spread of the disease during storage.

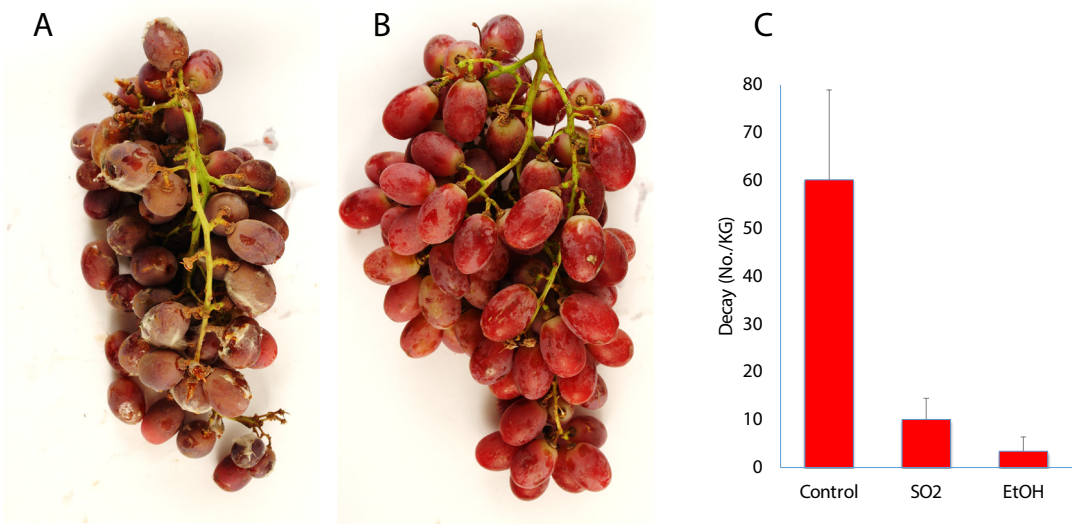

Figure 3: *The appearance of clusters of 'Scarlotta' grapes and the level of decay in the treatments after cold storage. A. Control untreated cluster. B. Ethanol (33%) treated cluster. C. The level of decay after 6 weeks of cold storage at 0°C and 3 days at 20°C. The grapes were stored in 9 kg boxes with SO_2 sheets above them. Ethanol was applied by dipping in a solution of 33% and drying for one hour. The boxes were wrapped with low density polyethylene during storage.*

.....reduction in the level of cracking by application of gibberellic acid during late fruit set can affect the level of postharvest decay

Conclusions

The results demonstrate that reduction in the level of cracking by application of gibberellic acid during late fruit set can affect the level of postharvest decay. This reduction in cracking is not sufficient to prevent postharvest decay by itself but when combined with other approaches it can allow the use of milder postharvest treatments. Additionally, disinfection of table grapes after harvest can have a dramatic effect on prevention of decay in grapes. This approach can be implemented in several ways but it also has an advantage by removing dirt that accumulates on the berries throughout the season. These two approaches are part of a 'multiple hurdle approach' which takes into account that there is no single step or protocol that can allow full protection against postharvest losses and that protection against postharvest losses is possible during all stages of fruit development.

References

[1] Lavee, S. (1960) Effect of gibberellic acid on seeded grapes. Nature 185, 395.

[2] Lichter, A., F.M. Gabler, and J.L. Smilanick (2006) Control of spoilage in table grapes. Stewart Postharvest Review 2, 1-10.

[3] Opera, L.U., C.I. Studman, and N. Banks (1996) Fruit skin splitting and cracking. Hort. Rev 19, 217-Z262.

[4] Reynolds, A., D. Wardle, C. Zurowski, and N. Looney (1992) Phenylureas CPPU and thidiazuron affect yield components, fruit composition, and storage potential of four seedless grape selections. Journal of the American Society for Horticultural Science 117, 85-89.

About the main Author

Dr Amnon Lichter is scientist in the Department of Postharvest Sciences since 1998 and served as the chair of the department from 2010 to 2013. Table grapes are the major focus in the current research program of Dr Lichter and it includes a wide range of activities: understanding the effects of pre-harvest developmental and horticultural factors on the postharvest quality; search for new varieties with improved postharvest properties; prevention of decay by *Botrytis cinerea* during storage; stem browning during storage; adoption of new technologies for phenotyping postharvest traits of grapes including image analysis and auto-fluorescence. A major part of the current program is better understanding of flavor in grapes from the perspective of tannins, volatile compounds and how we can maintain and improve unique flavor components. Past research also included abscission in bunch tomatoes, postharvest of litchi fruit, sanitation technologies and involvement in diverse postharvest research programs.

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