36

Future Studies on Postharvest Physiological Disorders in Fruit and Vegetables

Sergio Tonetto de Freitas and Sunil Pareek

CONTENTS

List of Abbreviations

CO_2	Carbon dioxide
CRISPR	Clustered regularly interspaced short palindromic repeats
CRISPR/Cas	CRISPR-associated system
CT	X-ray computed tomography
FTIR	Fourier transform infrared
gRNA	Guide RNA
HIS	Hyperspectral imaging
HSPs	Heat shock proteins
NIR	Near-infrared
NIRS	Near-infrared spectroscopy
O_2	Oxygen
PPO	Polyphenol oxidase
ROS	Reactive oxygen species

36.1 Introduction

The increasing world population and food quality standards necessitate a better understanding of the mechanisms regulating physiological disorders in different crop species to improve crop production efficiency by reducing losses. Although many physiological disorders have been studied and described in the literature, a larger number of disorders still remain poorly understood and very little information is available about the regulating mechanisms as well as possible approaches to predict and control the

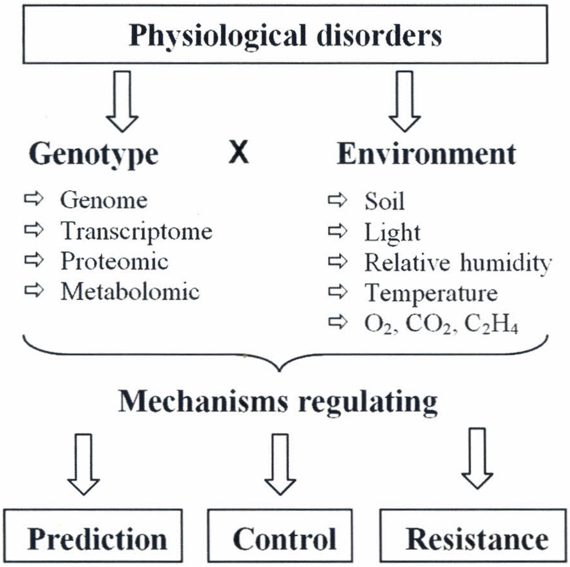

FIGURE 36.1 Proposed approach to study physiological disorders in fruits and vegetables.

incidence of physiological disorders. In addition, low precision in the description and identification of disorder symptoms can cause confusion because the same disorder can be described differently, or may not be the same disorder recognized by others. Misidentification of physiological disorders can lead to confusion in determining the mechanisms regulating their development in plants, emphasizing the importance of the precise characterization and identification of disorders.

The susceptibility of fruits and vegetables to disorders is determined by the interaction between genotype and environment (Figure 36.1). In that case, understanding the mechanisms regulating disorders requires knowledge of the genetic and environmental factors involved in the pathways leading to disorder development in each genotype. These factors can regulate crop susceptibility to disorders at the whole plant, tissue, and/or cellular levels.

36.2 Genotype

Since crop genotypes have different susceptibility to physiological disorders, studies have attempted to determine genetic markers that could be used to identify genotypes highly susceptible to disorders (Buti et al., 2015; Korban and Swiader, 1984; Kumar et al., 2013; McClure et al., 2016). However, other studies have shown that mechanisms regulating disorder incidence are highly complex and may not be fully explained by a few genetic markers. Indeed, physiological disorders can involve changes in both primary and secondary metabolism, including energy-related changes linked to the tricarboxylic acid cycle and the electron transport chain, oxidative stress, and lipid-related genes associated with membrane changes and fatty acid oxidation (Freitas et al., 2017; Lee et al., 2012; Leisso et al., 2013; Leisso et al., 2015; Leisso et al., 2016; Lum et al., 2016; Rudell et al., 2009). Considering the large number of pathways that may be involved, a detailed characterization of the mechanisms regulating physiological disorders can be accomplished using recently developed high-throughput approaches at the genomic, transcriptome, proteomic, and metabolomic levels (Figure 36.1). The information obtained can be used in systems biology approaches to facilitate a multi-targeted approach by allowing one to identify regulatory hubs in complex networks regulating physiological disorders in fruit and vegetables. Systems biology takes the molecular parts (transcripts, proteins, and metabolites) of each crop genotype and attempts to fit them into functional networks or models designed to describe and predict the dynamic activities of that genotype in different environments (Cramer et al., 2011). In that case, systems biology approaches can help in determining key regulatory mechanisms that can be used to predict, control, and/or develop plant genotypes that are more resistant to physiological disorders.

36.3 Environment

Since disorders are strongly affected by the environment, the role of environmental factors such as soil, light, relative humidity, temperature, O_2, CO_2, and ethylene on disorder development should be evaluated in susceptible genotypes (Figure 36.1). Although difficult to accomplish, the role of environmental factors on the mechanisms triggering or inhibiting disorder development in each genotype should be conjointly and independently evaluated. The knowledge obtained will help in determining more efficient pre-harvest and postharvest crop management practices to inhibit or reduce disorder incidence.

36.4 Developing Approaches to Predict Disorders

Many crop genotypes have high susceptibility to physiological disorders. In that case, developing efficient and precise approaches to predict disorder incidence can help to determine appropriate management practices to inhibit or control their incidence, as well as to predict crop postharvest life for commercialization. In addition, precise methods to predict disorder incidence can be used in approaches aiming to identify key regulatory mechanisms triggering or inhibiting disorders before the development of visual symptoms. Later, identified key regulatory mechanisms can be used as markers for the selection of new genotypes, as well as to improve the prediction efficiency of disorder incidence.

36.4.1 Modeling Crop Quality and Environmental Factors

Since physiological disorders are determined by genetic and environmental factors (Figure 36.1), statistical models can be developed to predict disorder incidence based on the range of the most important factors regulating each disorder. Indeed, studies have shown that specific genes, proteins, metabolites, and nutrients, as well as soil, light, relative humidity, temperature, and gas (O_2, CO_2) conditions, are highly correlated with disorder incidence. In these studies, some reported factors triggering physiological disorders are high levels of reactive oxygen species (ROS) and growth regulators (gibberellins and ethylene); low energy balance; high nitrogen, magnesium, and potassium contents; low calcium content; high membrane leakage; abnormal cell wall metabolism; high photosynthesis rate; high leaf/fruit ratio; high growth rate; loss of xylem functionality; drought; low soil nutrient availability; high or low light intensity; temperature; relative humidity; as well as low O_2 and high CO_2 levels during storage, among others (Amarante et al., 2013; Baugher et al., 2017; Freitas et al., 2010; Freitas et al., 2016; Freitas et al., 2017; Ho and White, 2005; Khadivi-Khub, 2015; Miqueloto et al., 2011; Nock and Watkins, 2013; Saquet and Streif, 2008; Saure, 2014). Other studies have shown that factors inhibiting physiological disorders are high antioxidant capacity, high levels of growth regulators (abscisic acid, gibberellins, and ethylene inhibitors), high calcium content, normal cell wall metabolism, and low sugar content, among others (Castro et al., 2007; Falchi et al., 2017; Freitas et al., 2012; Freitas et al., 2017; Lázaro and Lorenzo, 2014; Mckenzie et al., 2013). The information about factors triggering or inhibiting each disorder incidence can guide the development of efficient and precise statistical models to predict each disorder incidence in fruits and vegetables. Although a few studies have shown the possibility of predicting disorders (Amarante et al., 2013; Lotze et al., 2006a; Torres et al., 2017), the models developed are based on only a few nutritional variables, resulting in low model predictive performance and robustness. Considering the high number of variables regulating disorders, high model predictive performance and robustness can be achieved by developing multivariate models containing a larger number of variables that can increase model prediction efficiency in a wide range of genetic and environmental conditions.

36.4.2 Non-destructive Prediction Methods

The use of non-destructive methods to determine fruit and vegetable quality has increased in the last few years. Among these methods, near-infrared spectroscopy (NIRS) has proven to be a reliable analytical technique for qualitative and quantitative analyses, as well as for the prediction of physiological disorders in fruits and vegetables (Jarolmasjed et al., 2017; Nicolaï et al., 2009; Zúñiga et al., 2017). The technique has the advantage of being non-destructive, rapid, and precise, and also requires no sample preparation, which makes NIRS an important tool to predict disorders in different crop species. Studies have shown that near-infrared (NIR) spectral bands of 971.2, 978.0, 986.1, 987.3, 995.4, 1131.5, 1135.3, 1139.1, and 1142.8 nm are highly associated with bitter pit incidence in apples, and were used to distinguish pitted from non-pitted fruit with average accuracy in the range of 78–87% (Kafle et al., 2016). In addition, Fourier transform infrared (FTIR) spectroscopy has also been shown to predict with high accuracy physiological disorders in apples (Zúñiga et al., 2017). In both cases, NIRS and FTIR technologies are based on a single spectrum, usually an average spectrum, collected from a limited area on a fruit or vegetable. A more complex technology known as hyperspectral imaging (HSI) integrates conventional imaging, spectroscopy, and chemometrics to attain both spatial and spectral information from an object (Amigo et al., 2013; Gowen et al., 2007; Lorente et al., 2012). HIS has been shown to have high prediction performance in identifying common defects on the surface of fruits and vegetables, such as bruises and cracks on jujube (Wu et al., 2016), bitter pit not visible to the naked eye at harvest, black rot, decay, soft scald, superficial scald, chilling injury in apples (Ariana et al., 2006; ElMasry et al., 2009; Nicolaï et al., 2006), bloater damage in whole pickles, (Ariana and Lu, 2010), as well as internal and external defects in citrus fruit (Magwaza et al., 2012) and hidden bruises in kiwi fruit (Lü and Tang, 2012). Although HIS can help identify disorders in fruit and vegetables, future studies should focus on increasing HIS predictive performance and robustness to avoid prediction mistakes. For example, HSI has been shown to identify bitter pit lesions, but could not discriminate between a disorder's lesions and corky tissue (Nicolaï et al., 2006). Other studies have also shown that imaging systems may have low predictive performance (Lotze et al. 2006b; Mirzaee et al., 2015), which could limit their application in commercial conditions. X-ray computed tomography (CT) imaging is another non-destructive and rapid sensing technique that can be used to predict disorder incidence in fruits and vegetables. CT images have been shown to precisely predict external and internal bitter pits in apples during storage, which could be used to effectively identify this disorder in an automated manner under commercial conditions (Jarolmasjed et al., 2016; Si and Sankaran, 2016). Other studies have suggested that NIR tomography could be the natural progression of NIRS, offering the possibility of detecting external and internal defects in fruit and vegetables (Kemsley et al., 2008). Although the technologies presented here can be used to predict and monitor agricultural produce quality and disorders, future studies should focus on developing low-cost, portable, calibrated, and validated scanners for each genotype and growing condition to guarantee the high predictive performance and robustness of these technologies in commercial conditions.

36.4.3 Biomarkers

Although most disorders are regulated by complex mechanisms, biomarkers can help monitor and predict disorder incidence in fruit and vegetables. Indeed, studies have identified biomarkers as related to the resistance or susceptibility of fruits and vegetables to physiological disorders. For example, chilling injury has been attributed to membrane damage and ROS production, which are counteracted by heat shock proteins (HSPs) (Aghdam et al., 2013). Therefore, the analysis of HSPs could be envisaged as an ideal method for assessing fruit and vegetable resistance to chilling injury and for evaluating the efficiency of postharvest treatments on inhibiting chilling injury incidence (Aghdam et al., 2013). Similar to HSPs, other biomarkers have been shown to be correlated with higher or lower susceptibility to chilling injury (Aghdam and Bodbodak, 2013), bitter pit (Buti et al., 2015; Korban and Swiader, 1984; Kumar et al., 2013), soft scald (McClure et al., 2016), and blossom-end rot (Freitas et al., 2017), among others. After identifying the most precise biomarkers positively or negatively correlated to each disorder, the development of simple and precise biomarker quantification kits can help determine changes in fruit and vegetable susceptibility to disorders. This technology could be used in the field as well as during storage and commercialization to predict and monitor disorder incidence, which will help in determining specific management approaches to inhibit or reduce disorder incidence, as well as to properly plan fruit and vegetable commercialization based on the postharvest lifespan of specific produce. Additionally, such tools could also be applied to diagnose disorder incidence after it has occurred.

36.5 Developing Management Approaches to Control Disorders

Based on the approach used to predict disorder incidence, fruit and vegetables presenting high susceptibility should be properly managed to reduce or inhibit disorder incidence, thus reducing crop losses. The most recommended approaches to reduce or inhibit each disorder incidence have been described in detail in the previous chapters. In addition, future studies looking at disorder control approaches will add information and alternatives to control different disorders in fruit and vegetables. Alternatively, fruit and vegetables showing high susceptibility to disorder incidence after harvest should be marketed faster to guarantee consumption before the appearance of disorders.

36.6 Developing Resistant Genotypes to Disorders

The knowledge about genomic, transcriptome, proteomic, and metabolomic mechanisms regulating physiological disorders can be used to identify key regulatory genes and genetic markers that can guide breeding programs and genome transformation and editing approaches to obtain highly resistant plants (Figure 36.1). For example, proteomic and transcriptional studies have identified candidate genes that trigger or inhibit the disorder known as blossom-end rot in tomato fruit (Casado-Vela et al., 2005; Freitas et al., 2017). The proteomic study shows the induction of proteins participating in antioxidant processes such as the ascorbate-glutathione cycle and the pentose phosphate pathway in affected fruit, suggesting that these two biochemical pathways, acting as ROS scavengers, restrain the spread of the disorder in the fruit (Casado-Vela et al., 2005). The transcriptome study shows that genes inhibiting blossom-end rot in tomato have functions leading to higher resistance to oxidative stress and toxic compounds, whereas most of the candidate genes triggering the disorder have functions leading to higher levels of oxidative stress and cell death (Freitas et al., 2017). Based on these findings, resistant genotypes to blossom-end rot could be selected for a higher expression of genes coding for enzymes involved in antioxidant metabolism, as well as a lower expression of genes involved in oxidative stress and cell death. Alternatively, genome transformation and editing techniques could be used to silence genes triggering and/or increasing the expression of genes inhibiting blossom-end rot in tomato fruit. In another example, studies have shown that polyphenol oxidase (PPO) catalyzes the oxidation of phenolic compounds into highly reactive quinones, which causes tissue browning in fruit and vegetables (Araji et al., 2014; Waltz, 2015). Considering that tissue browning due to oxidation is one of the most common symptoms of many physiological disorders, silencing PPO expression can have a great effect on alleviating disorder symptom development. Indeed, PPO-silenced potatoes and apples have been shown to develop much lower browning pigments than wild types (Chi et al., 2014; Waltz, 2015). However, the effect of PPO silencing on disorders development should be investigated because PPO-silenced plants can have higher tyramine content, which is known to elicit cell death, suggesting that PPO may play a fundamental role in secondary metabolism and as a regulator of cell death (Araji et al., 2014). Other candidate genes and genetic markers that can potentially be used to develop resistant genotypes have been describe in the literature and many more will be identified in future studies (Capel et al., 2017; Cavaiuolo et al., 2015; Ikeda et al., 2017; Khadivi-Khub, 2015; Martínez et al., 2009; Saeed et al., 2014; Sanchez-Ballesta et al., 2008).

Although breeding programs have been extensively used to obtain new crop genotypes with desired features, targeted genome editing using artificial nucleases can accelerate the development of new genotypes by providing the means to modify genomes rapidly in a precise and predictable manner (Bortesi and Fisher, 2015). Among genome editing platforms are the well-established zinc finger nucleases and transcription activator-like effector nucleases, and the recently developed clustered regularly interspaced short palindromic repeats (CRISPR) (Bortesi and Fisher, 2015). CRISPR/Cas (CRISPR-associated system) is a rapidly developing genome editing technology, and its simple version known as CRISPR/Cas9 has been modified to edit genomes. By delivering the Cas9 nuclease complexed with a synthetic guide RNA (gRNA) into a cell, the cell's genome can be modified at a desired location, silencing or stimulating the expression of specific genes (Bortesi and Fisher, 2015; Ledford, 2015, 2016). As only a short RNA sequence must be synthesized to confer recognition of a new target, CRISPR/Cas9 is a relatively cheap and easy to implement technology that has proven to be extremely versatile (Belhaj et al., 2015; Bortesi and Fisher, 2015). Therefore, sequence-specific nucleases, such as CRISPR/Cas9-gRNA complex, are powerful tools for genome editing (Ledford, 2015, 2016) that can be used to make the specific genome changes required to solve specific problems such as physiological disorders in plants.

Therefore, the knowledge obtained about the mechanisms regulating physiological disorders in plants can be used to develop efficient and precise approaches to predict, control, as well as obtain new genotypes more resistant to disorders through breeding programs and/or recently developed genome transformation and editing techniques (Figure 36.1). Mechanisms regulating physiological disorders in studied crop species will help us to understand the mechanisms regulating disorders in less-studied plant genotypes.

REFERENCES

Aghdam, M.S. and Bodbodak, S. 2013. Physiological and biochemical mechanisms regulating chilling tolerance in fruits and vegetables under postharvest salicylates and jasmonates treatments. *Scientia Horticulturae* 156, 73–85.

Aghdam, M.S., Sevillano, L., Flores, F.B., and Bodbodak, S. 2013. Heat shock proteins as biochemical markers for postharvest chilling stress in fruits and vegetables. *Scientia Horticulturae* 160, 54–64.

Amarante, C.V.T., Miqueloto, A., Freitas, S.T., Steffens, C.A., Silveira, J.P.G., and Corrêa, T.R. 2013. Fruit sampling methods to quantify calcium and magnesium contents to predict bitter pit development in "Fuji" apple: A multivariate approach. *Scientia Horticulturae* 157, 19–23.

Amigo, J.M., Martí, I., and Gowen, A. 2013. Hyperspectral imaging and chemometrics. *Data Handling in Science and Technology* 28, 343–370.

Araji, S., Grammer, T.A., Gertzen, R., Anderson, S.D., Mikulic-Petkovsek, M., Veberic, R., Phu, M.L., et al. 2014. Novel roles for the polyphenol oxidase enzyme in secondary metabolism and the regulation of cell death in walnut. *Plant Physiology* 164, 1191–1203.

Ariana, D.P. and Lu, R. 2010. Evaluation of internal defect and surface color of whole pickles using hyperspectral imaging. *Journal of Food Engineering* 96, 583–590.

Ariana, D., Guyer, D.E., and Sherstha, B. 2006. Integrating multispectral reflectance and fluorescence imaging for defect detection on apples. *Computers and Electronics in Agriculture* 50, 148–161.

Baugher, T.A., Marini, R., Schupp, J.R., and Watkins, C.B. 2017. Prediction of bitter pit in "Honeycrisp" apples and best management implications. *Hortscience* 52, 1368–1374.

Belhaj, K., Chaparro-Garcia, A., Kamoun, S., Patron, N.J., and Nekrasov, V. 2015. Editing plant genomes with CRISPR/Cas9. *Current Opinion in Biotechnology* 32, 76–84.

Bortesi, L. and Fisher, R. 2015. The CRISPR/Cas9 system for plant genome editing and beyond. *Biotechnology Advances* 33, 41–52.

Buti, M., Poles, L., Caset, D., Magnago, P., Fernandez, F., Colgan, R.J., Velasco, R., and Sargent, D.J. 2015. Identification and validation of a QTL influencing bitter pit symptoms in apple (*Malus x domestica). Molecular Breeding* 35, 1–11.

Capel, C., Yuste-Lisbona, F.J., López-Casado, G., Angosto, T., Cuartero, J., Lozano, R., and Carpel, J. 2017. Multi-environment QTL mapping reveals genetic architecture of fruit cracking in a tomato RIL *Solanum lycopersicum×S. pimpinellifolium* population. *TAG. Theoretical and Applied Genetics. Theoretische und Angewandte Genetik* 130, 213–222.

Casado-Vela, J., Sellés, S., and Martínez, R.B. 2005. Proteomic approach to blossom-end rot in tomato fruits (*Lycopersicon esculentum* M.): Antioxidant enzymes and the pentose phosphate pathway. *Proteomics* 5, 2488–2496.

Castro, E., Biasi, B., Mitcham, E., Tustin, S., Tanner, D., and Jobling, J. 2007. Carbon dioxide-induced flesh browning in Pink Lady apples. *Journal of the American Society for Horticultural Science* 132, 713–719.

Cavaiuolo, M., Cocetta, G., Bulgari, R., Spinardi, A., and Ferrante, A. 2015. Identification of innovative potential quality markers in rocket and melon fresh-cut produce. *Food Chemistry* 188, 225–233.

Chi, M., Bhagwat, B., Lane, W.D., Tang, G., Su, Y., Sun, R., Oomah, B.D., Wiersma, P.A., and Xiang, Y. 2014. Reduced polyphenol oxidase gene expression and enzymatic browning in potato (*Solanum tuberosum* L.) with artificial microRNAs. *BMC Plant Biology* 14, 62.

Cramer, G.R., Urano, K., Delrot, S., Pezzotti, M., and Shinozaki, K. 2011. Effects of abiotic stress on plants: A systems biology perspective. *BMC Plant Biology*, 163.

ElMasry, G., Wang, N., and Vigneault, C. 2009. Detecting chilling injury in red delicious apple using hyperspectral imaging and neural networks. *Postharvest Biology and Technology* 52, 1–8.

Falchi, R., D'Agostin, E., Mattiello, A., Coronica, L., Spinelli, F., Costa, G., and Vizzotto, G. 2017. ABA regulation of calcium-related genes and bitter pit in apple. *Postharvest Biology and Technology* 132, 1–6.

Freitas, S.T., Martinelli, F., Feng, B., Reitz, N.F., and Mitcham, E.J. 2017. Transcriptome approach to understand the potential mechanisms inhibiting or triggering blossom-end rot development in tomato fruit in response to plant growth regulators. *Journal of Plant Growth Regulation* 37, 183–198.

Freitas, S.T., Amarante, C.V.T., and Mitcham, E.J. 2016. Calcium deficiency disorders in plants. In S. Pareek (Ed.), *Postharvest Ripening Physiology of Crops*, 477–512. CRC Press: Boca Raton, FL.

Freitas, S.T., Pereira, E.I.P., Gomez, A.C.S., Brackmann, A., Nicoloco, F., and Bisognin, D.A. 2012. Processing quality of potato tubers produced during autumn and spring and stored at different temperatures. *Horticultura Brasileira* 30, 91–98.

Freitas, S.T., Amarante, C.V.T., Labavitch, J.M., and Mitcham, E.J. 2010. Cellular approach to understand bitter pit development in apple fruit. *Postharvest Biology and Technology* 57, 6–13.

Gowen, A.A., O'Donnell, C.P., Cullen, P.J., Downey, G., and Frias, J.M. 2007. Hyperspectral imaging – An emerging process analytical tool for food quality and safety control. *Trends in Food Sciences & Technology* 18, 590–598.

Ho, L.C. and White, P.J. 2005. A cellular hypothesis for the induction of blossom-end rot in tomato fruit. *Annals of Botany* 95, 571–581.

Ikeda, H., Shibuya, T., Nishiyama, M., Nakata, Y., and Kanayama, Y. 2017. Physiological mechanisms accounting for the lower incidence of blossom-end rot in tomato introgression line IL8-3 fruit. *The Horticulture Journal* 86, 327–333.

Jarolmasjed, S., Espinoza, C.Z., and Sankaran, S. 2017. Near infrared spectroscopy to predict bitter pit development in different varieties of apples. *Journal of Food Measurement and Characterization* 11, 987–993.

Jarolmasjed, S., Espinoza, C.Z., Sankaran, S., and Khot, L.R. 2016. Postharvest bitter pit detection and progression evaluation in 'Honeycrisp' apples using computed tomography images. *Postharvest Biology and Technology* 118, 35–42.

Kafle, G.K., Khot, L.R., Jarolmasjed, S., Yongsheng, S., and Lewis, K. 2016. Robustness of near infrared spectroscopy based spectral features for non-destructive bitter pit detection in Honeycrisp apples. *Postharvest Biology and Technology* 120, 188–192.

Kemsley, E.K., Tapp, H.S., Binns, R., Mackin, R.O., and Peyton, A.J. 2008. Feasibility study of NIR diffuse optical tomography on agricultural produce. *Postharvest Biology and Technology* 48, 223–230.

Khadivi-Khub, A. 2015. Physiological and genetic factors influencing fruit cracking. *Acta Physiologiae Plantarum* 37, 1718.

Korban, S.S. and Swiader, J.M. 1984. Genetic and nutritional status in bitter pit-resistant and pit-susceptible apple seedlings. *Journal of the American Society for Horticultural Science* 109, 428–432.

Kumar, S., Garrick, D.J., Bink, M.C., Whitworth, C., Chagne, D., and Volz, R.K. 2013. Novel genomic approaches unravel genetic architecture of complex traits in apple. *BMC Genomics* 14, 393.

Lázaro, A. and Lorenzo, C. 2014. Texture analysis in melon landraces through instrumental and sensory methods. *International Journal of Food Properties* 18, 1575–1583.

Ledford, H. 2015. CRISPR, the disruptor. *Nature* 522, 20–24.

Lee, J., Cheng, L., Rudell, D.R., and Watkins, C.B. 2012. Antioxidant metabolism of 1-methylcyclopropene (1-MCP) treated "Empire" apples during controlled atmosphere storage. *Postharvest Biology and Technology* 65, 79–91.

Ledford, H. 2016. CRISPR: Gene editing is just the beginning. *Nature* 531, 156–159.

Leisso, R.S., Gapper, N.E., Mattheis, J.P., Sullivan, N.L., Watkins, C.B., Giovannoni, J.J., Schaffer, R.J., et al. 2016. Gene expression and metabolism preceding soft scald, a chilling injury of "Honeycrisp" apple fruit. *BMC Genomics* 17.

Leisso, R.S., Buchanan, D.A., Lee, J., Mattheis, J.P., Sater, C., Hanrahan, I., Watkins, C.B., et al. 2015. Chilling-related cell damage of apple (*Malus x domestica* Borkh.) fruit cortical tissue impacts antioxidant, lipid and phenolic metabolism. *Physiologia Plantarum* 153, 204–220.

Leisso, R., Buchanan, D., Lee, J., Mattheis, J., and Rudell, D. 2013. Cell wall, cell membrane, and volatile metabolism are altered by antioxidant treatment, temperature shifts, and peel necrosis during apple fruit storage. *Journal of Agricultural and Food Chemistry* 61, 1373–1387.

Lorente, D., Aleixos, N., Gómez-Sanchis, J., Cubero, S., García-Navarrete, O.L., and Blasco, J. 2012. Recent advances and applications of hyperspectral imaging for fruit and vegetable quality assessment. *Food and Bioprocess Technology* 5, 1121–1142.

Lotze, E., Sadie, A., and Theron, K.I. 2006a. Determining the probability of bitter pit in "golden Delicious" apples through the postharvest mineral content of individual fruit. *Journal of Horticultural Science & Biotechnology* 81, 276–280.

Lotze, E., Huybrechts, C., Sadie, A., Theron, K.I., and Valcke, R.M. 2006b. Fluorescence imaging as a non-destructive method for pre-harvest detection of bitter pit in apple fruit (*Malus domestica* Borkh.). *Postharvest Biology and Technology* 40, 287–294.

Lü, Q. and Tang, M. 2012. Detection of hidden bruise on kiwi fruit using hyperspectral imaging and parallelepiped classification. *Procedia Environmental Sciences* 12, 1172–1179.

Lum, G.B., Brikis, C.J., Deyman, K.L., Subedi, S., DeEll, J.R., Shelp, B.J., and Bozzo, G.G. 2016. Pre-storage conditioning ameliorates the negative impact of 1-methylcyclopropene on physiological injury and modifies the response of antioxidants and y-aminobutyrate in "Honeycrisp" apples exposed to controlled-atmosphere conditions. *Postharvest Biology and Technology* 116, 115–128.

Magwaza, L.S., Opara, U.L., Nieuwoudt, H., Cronje, P.J.R., Saeys, W., and Nicolaï, B. 2012. NIR spectroscopy applications for internal and external quality analysis of citrus fruit – A review. *Food and Bioprocess Technology* 5, 425–444.

Martínez, J.A., Jowkar, M.M., Obando-Ulloa, J.M., Varó, P., Moreno, E., Monforte, A.J., and Fernández-Trujillo, J.P. 2009. Uncommon disorders and decay in near-isogenic lines of melon and reference cultivars. *Horticultura Brasileira* 27, 505–514.

McClure, K.A., Gardner, K.M., Toivonen, P.M.A., Hampson, C.R., Song, J., Forney, C.F., DeLong, J., Rajcan, I., and Myles, S. 2016. QTL analysis of soft scald in two apple populations. *Horticulture Research* 3, 16043.

McKenzie, M.J., Chen, R.K.Y., Harris, J.C., Ashworth, M.J., and Brummell, D.A. 2013. Post-translational regulation of acid invertase activity by vacuolar invertase inhibitor affects resistance to cold-induced sweetening of potato tubers. *Plant, Cell and Environment* 36, 176–185.

Miqueloto, A., Amarante, C.V.T., Steffens, C.A., Santos, A., Miqueloto, T., and Silveira, J.P.G. 2011. Physiological, physicochemical and mineral attributes associated with the occurrence of bitter pit in apples. *Pesquisa Agropecuária Brasileira* 46, 689–696.

Mirzaee, M., Rees, D., Colgan, R.J., and Tully, M.S. 2015. Diagnosing bitter pit in apple during storage by chlorophyll fluorescence as a non-destructive tool. *Acta Horticulturae* 1079, 235–242.

Nicolaï, B.M., Bulens, I., De Baerdemaker, J., De Ketelaere, B., Hertog, M.L.A.T.M., Verboven, P., and Lammertyn, J. 2009. Non-destructive evaluation: Detection of external and internal attributes frequently associated with quality and damage. In W. J. Florkowski, R.L. Shewfelt, B. Brueckner, S.E. Prussia (Eds.), *Postharvest Handling: A Systems Approach*, 421–442. Academic Press, Elsevier: Amsterdam.

Nicolaï, B.M., Lotze, E., Peirs, A., Scheerlinck, N., and Theron, K.I. 2006. Non-destructive measurement of bitter pit in apple fruit using NIR hyperspectral imaging. *Postharvest Biology and Technology* 40, 1–6.

Nock, J.F. and Watkins, C.B. 2013. Repeated treatment of apple fruit with 1-methylcyclopropene (1-MCP) prior to controlled atmosphere storage. *Postharvest Biology and Technology* 79, 73–79.

Rudell, D.R., Mattheis, J.P., and Hertog, M.L. 2009. Metabolomic change precedes apple superficial scald symptoms. *Journal of Agricultural and Food Chemistry* 57, 8459–8466.

Saeed, M., Brewer, L., Johnston, J., McGhie, T.K., Gardiner, S.E., Heyes, J.A., and Chagné, D. 2014. Genetic, metabolite and developmental determinism of fruit friction discolouration in pear. *BMC Plant Biology* 14, 241.

Saquet, A.A. and Streif, J. 2008. Fermentative metabolism in "Jonagold" apples under controlled atmosphere storage. *European Journal of Horticultural Science* 73, 43–46.

Sanchez-Ballesta, M.T., Zacarías, L., Granell, A., and Lafuente, M.T. 2008. ß-1,3-glucanase gene expression as a molecular marker for postharvest physiological disorders in citrus fruit and its hormonal regulation. *Postharvest Biology and Technology* 48, 146–149.

Saure, M.C. 2014. Why calcium deficiency is not the cause of blossom-end rot in tomato and pepper fruit – A reappraisal. *Scientia Horticulturae* 174, 151–154.

Si, Y.S. and Sankaran, S. 2016. Computed tomography imaging-based bitter pit evaluation in apples. *Biosystems Engineering* 151, 9–16.

Torres, E., Recasens, I., Avila, G., Lordan, J., and Alegre, S. 2017. Early stage fruit analysis to detect a high risk of bitter pit in "Golden Smoothee". *Scientia Horticulturae* 219, 98–106.

Waltz, E. 2015. Nonbrowning GM apple cleared for Market. *Nature Biotechnology* 33, 326–327.

Wu, L., He, J., Liu, G., Wang, S., and He, X. 2016. Detection of common defects on jujube using vis-NIR and NIR hyperspectral imaging. *Postharvest Biology and Technology* 112, 134–142.

Zúñiga, C.E., Jarolmasjed, S., Sinha, R., Zhang, C., Kalcsits, L., Dhingra, A., and Sankaran, S. 2017. Spectrometric techniques for elemental profile analysis associated with bitter pit in apples. *Postharvest Biology and Technology* 128, 121–129.