Effects of Vacuum Packaging on Enzymatic Browning and Ethylene Response Factor (ERF) Gene Expression of Fresh-cut Lotus Root

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Additional index words. Nelumbo nucifera, browning, PAL, PPO, POD, ERF

Abstract. Ethylene response factor (ERF) genes have been involved in responses to biotic and abiotic stress, including hypoxia and anaerobic stress. Vacuum packaging (a typical anaerobic stress) is an effective storage method used to delay browning of fresh-cut lotus root (Nelumbo nucifera). In model plants, ERF genes have been identified as responsive to hypoxia. Whether ERF is associated with browning of vacuum-packaged lotus root has not been studied. The effects of vacuum packaging on browning, phenolic content, the enzyme activity of phenylalanine ammonia lyase (PAL), polyphenol oxidase (PPO), and peroxidase (POD), and PPO, PAL, POD, and ERF genes expression in fresh-cut lotus root were studied. Downregulation of NnPAL1, NnPPO4, and NnPPO2/3 attributable to vacuum packaging coincided with increased related enzyme activities and the degree of browning of fresh-cut lotus root. The expression patterns of NnERF4/5 were consistent with the changes in NnPAL1, NnPPO4, and NnPPO2/3 gene expression. It has been proposed that NnERF4/5 could have be important regulators of fresh-cut lotus root browning, and that the relationships of NnPAL1, NnPPO4, and NnPPO2/3 should to be studied further.

Lotus root is an important aquatic vegetable and export vegetable in China. There are abundant nutrients and high nutritional value, mainly including starch, sugar, protein, fat, and lecithin in lotus root (Solina-Fortuny and Martin-Belloso, 2003). Because it is easy to cut, lotus root is more suitable for processing into fresh-cut products (Du et al., 2009). Fresh-cut lotus root is a fast and convenient food, and its processing procedure includes grading, cleaning, cutting, rest, preservation, and packaging. However, during processing and storage, fresh-cut lotus root is highly susceptible to deterioration, which directly affects its quality and shelf life. The main reason for the decline in quality of fresh-cut products has been enzymatic browning (Eissa et al., 2006; Pma, 2006; Son et al., 2015).

At present, the methods for controlling the browning of lotus root mainly include chemical treatment (Kwon and Baek, 2014; Lu et al., 2007), modified atmosphere (MA) packaging (Cheng et al., 2015), vacuum packaging (Son et al., 2015; Xing et al., 2012), heat treatment (Tsouvaltzis et al., 2011), and low-temperature storage (Min et al., 2017). Among these, vacuum packaging is a natural preservation method that can greatly enhance the shelf life and overall quality of fresh produce (Mcdonald and Sun, 2000). Phenylalanine ammonia lyase (PAL), polyphenol oxidase (PPO), and peroxidase (POD) are involved in the enzymatic browning of many fruits and vegetables (Banerjee et al., 2015; Cheng et al., 2015; Li-Lian et al., 2016; Zhou et al., 2003). However, vacuum packaging can delay the browning of fresh-cut lotus roots, and samples that have been vacuum-packaged have exhibited lower total phenol contents and PPO activity as well as lower degrees of browning than samples that have been air-packaged (Son et al., 2015; Xing et al., 2012). Our previous research reported that NnPAL1, NnPPO4, and NnPPO2/3 are the most important candidate genes involved in the browning of fresh-cut lotus root (‘E Liu 6’) during storage at different temperatures (Min et al., 2017). However, the effect of vacuum packaging on PAL, PPO, and POD genes have not been reported for fresh-cut lotus root.

Ethylene response factor (ERF) genes have been characterized in numerous plants in which they are involved in responses to biotic and abiotic stress, including hypoxia stress, cold stress, and heat stress (Licaisi et al., 2013; Min et al., 2012; M€uller and Munne-Bosch, 2015; Phukan et al., 2017). It has been reported that hypoxia-responsive ERF genes (HRE) have been characterized in Arabidopsis, with HRE1 and HRE2 having partially redundant roles in increasing low-oxygen tolerance (Hinz et al., 2010; Licaisi et al., 2010; Yang et al., 2011). Two hypoxia-responsive ERF genes (DkERF9 and DkERFI0) were involved in regulating persimmon deasstringency by separately regulating DkPDC2 and DkADH1 promoters (Min et al., 2012). Although the anaerobic environment produced by vacuum packaging or low temperatures is a type of unfavorable stress for plants, it has a remarkable effect on delaying browning of fresh-cut lotus root. In our previous study, we explored the role of ERF in delaying lotus root browning at low temperatures, and we found that NnERF3/4/5 could be important regulators of browning of fresh-cut lotus root during storage at low temperatures. Whether ERF is associated with the browning of lotus roots that are vacuum-packaged requires further study.

Ethylene can enhance rachis browning in Vitis vinifera and accelerate the browning of fresh-cut Colocasia esculenta during storage (Li and Zhang, 2015; Tan and Zeng, 2014). It is well known that ERF plays an important role in the realization of ethylene function (Nakano et al., 2006), suggesting that ERF may be involved in plant browning. ERF generally realizes functions by combining downstream genes. It has been reported that DcERF1 and DcERF2 may commit to the upregulation of DcPAL3 promoter activity in anhydrocyan-synthesizing carrot cells (Kimura et al., 2008). GhERF1 regulated lignin biosynthesis-related enzymes, including PAL, to improve resistance to Verticillium dahliae (Guo et al., 2016). A study of aphid resistance indicated that downregulation of ERF gene expression was consistent with the lower POD and PPO activities due to high CO2 (Guo et al., 2013). ThCRF1 (a subfamily member of the ERF transcription factor from Tamarix hispida) has improved
tolerance to salt-shock-induced stress by improving SOD and POD activities (Qin et al., 2017). This suggests that ERF may be involved in the synthesis of phenolic precursors through the transcriptional regulation of PAL in plants or the transcriptional regulation of PPO and POD involved in plant stress responses. Therefore, it is necessary to further study how ERF participates in the browning of lotus root that has been vacuum-packaged.

In this study, the effects of vacuum packaging on browning, total phenol, the enzyme activity of PPO, PAL, and POD, and the expression patterns of PPO, PAL, POD, and ERF were analyzed. Some PPO, PAL, POD, and NnERF genes were positively correlated with lotus root browning, and the possible roles of these and other NnERF genes are discussed.

Materials and Methods

Sample preparation

Lotus root (‘E lian 5’) was purchased from the commercial agricultural wholesale market (Four Seasons) in 2016; then, it was immediately shipped to the laboratory. It had a uniform size and color, and there were no defects or mechanical damage. Before samples were processed, the lotus roots were stored at 4 °C for 24 h. Then, lotus root was rinsed with clean water to remove the soil and peeled. Next, using a stainless-steel knife, it was cut into 5-mm-thick slices along the cross-section. The slices were transferred to clean water, sterilized by ozone for 5 min, and dried with clean filter paper. Some of the cymbals were vacuum-packed, and the other part was packaged with atmospheric pressure. Finally, samples were transferred to a refrigerator (4 °C).

Physicochemical parameters

Physicochemical parameters of fresh-cut lotus root slices, including the degree of browning and total phenolic content, were measured according to a previously published method (Min et al., 2017). All treatments were performed with three biological replicates.

Determination of the degree of browning

The degree of browning (Min et al., 2017) was determined using modified versions of methods referred to in the previous literature. At 4 °C, 30 mL of distilled water was mixed with 3.0 g of sliced lotus root tissue. It was homogenized and centrifuged for 5 min at 10,000 g. Next, the supernatant in the centrifuge tube was collected. After incubating for 5 min in a 25 °C water bath, the absorbance was measured at 410 nm using a spectrophotometer. The browning degree was expressed as A410.

Determination of the total phenolic content

The total phenolic content was measured according to the Folin-Ciocalteu method (Liu et al., 2018; Min et al., 2017). Three grams of sliced fresh lotus root tissue samples were homogenized with 30 mL of 60% ethanol and centrifuged at 10,000 g, for 5 min. The supernatant (10 mL) was diluted with 40 mL of 60% ethanol for the next measurement. The sample solution (0.125 mL) was mixed with 0.625 mL of distilled water; then, 0.125 mL of Folin phenol reagent was added. After thorough mixing (oscillation for 30 s), the mixture was permitted to stand at room temperature for 3 min; then, 1.25 mL of 7% Na2CO3 and 1.0 mL of distilled water were added. Finally, it was allowed to stand in a water bath at 25 °C for 90 min. Then, the absorbance was measured using a spectrophotometer at 760 nm. The standard curve of gallic acid was used to determine the total phenolic content. The result was expressed as milligrams of gallic acid equivalents per kilogram of fresh weight (mg·kg–1).

Determination of PPO, PAL, and POD enzyme activities

PAL activity was extracted and determined using a phenylalanine ammonia lyase activity assay kit (Nan Jing Jiancheng Bioengineering Institute, Nanjing, China) (Min et al., 2017). There were four reagents in the kit (reagents 1–4); 0.1 g of the sliced tissue was added to 1 mL of reagent 1 under ice bath conditions for homogenization. Then, the sample was centrifuged at 10,000 g for 10 min at 4 °C, and the supernatant was collected as a crude PAL extract. The reaction mixture consisted of 20 µL of crude PAL extract, 780 µL of reagent 2 (blank group 800 µL), and 200 µL of reagent 3. Afterward, the mixture was incubated at 30 °C for 30 min, and 40 µL of reagent 4 was added. Finally, it was allowed to stand at room temperature for 10 min, and the absorbance of the sample was measured at 290 nm. One unit of PAL activity was defined as the amount of enzyme that catalyzed when the change in absorbance was 0.1 per gram of fresh sample at 290 nm.

Extraction and determination of POD activity were based on the literature (Min et al., 2017). Under ice bath conditions, 3.0 g of sliced lotus root was homogenized in 50 mL of phosphate-buffered saline (PBS) (0.05 mol·L–1; pH = 7.0), and the supernatant was collected as a crude POD extract. Next, we prepared a reaction mixture consisting of 1.0 mL of 0.1 mol·L–1 catechol and 1.5 mL of PBS. The mixture was incubated at 35 °C for 5 min; then, 1.0 mL of crude POD was added to the mixture. Afterward, the absorbances of the mixture were measured at 420 nm over time. After the enzyme solution was added, the absorbance value was measured every 10 s during the first 1 min, and then every 30 s until 3.5 min. The absorbance value (OD) was measured over time. Finally, the enzyme activity was calculated based on the slope (OD/t) of the straight segment. An enzyme activity unit was described as the amount of enzyme leading to a change in absorbance of 0.001 per min.

Expression analysis of the ERF gene

Oligonucleotide primers for polymerase chain reaction (PCR) analyses were described previously (Min et al., 2015). According to previous literature, gene expression studies used the SsOvaFast EvaGreen Supermix kit (Bio-Rad) and CFX96 fluorescent PCR detection system for real-time PCR (Yang et al., 2018). Furthermore, 5.0 µL 2× SsoFast EvaGreen Supermix, 0.5 µL upstream primer (10 µmol), 0.5 µL downstream primer (10 µmol), 1.0 µL diluted cDNA, and 3.0 µL H2O were used.

The PCR procedure was as follows: 95 °C for 3 min; 95 °C for 10 s for 45 cycles; and 60 °C for 30 s. Then, the melting curve analysis was completed. The relative expression of each gene was calibrated using the sample from the day 0 control group. In addition, the relative expression levels of the genes were referenced to the lotus root Actin gene, and each gene was set as a negative control with no template. Each gene expression experiment was performed in triplicate.

Data analysis

Figures were drawn using Origin 8.0 software. Statistical analyses of differences was analyzed using least significant difference.

Results and Discussion

Browning degree

The browning degree was the main indicator of the browning of fruits and vegetables (Baxter, 2010). In this study, different packaging conditions (vacuum packaging and atmospheric packaging at 4 °C) were used to store fresh-cut lotus root slices. The results showed that the browning degree during storage was significantly different between the two packaging methods (Fig. 1). Compared with atmospheric packaging, the browning degree of fresh-cut lotus roots in vacuum packaging was markedly delayed (between 0.215 and 0.285), and the browning degree of roots in atmospheric packaging
changed greatly (between 0.215 and 0.610). Therefore, vacuum packaging could effectively delay the browning process of fresh-cut lotus root; these results were similar to the results of a previous study (Shan et al., 2013).

**Total phenolic content**

As shown in Fig. 2, the total phenolic content of fresh-cut lotus roots was significantly different in various packaging methods. The total phenolic content of the vacuum-packaged sample at 4 °C decreased and then increased, but the content was not significantly different from beginning to end (96.891 mg/kg on day 0 to 108.981 mg/kg on day 35). The total phenolic content of atmospheric pressure-packaged samples increased greatly from 96.891 mg/kg on day 0 to 159.252 mg/kg on day 35. Moreover, the results showed that storage in vacuum packaging delayed the increase in the total phenolic content in fresh-cut slices; these results were consistent with those of a previous study (Min et al., 2017).

**PAL, PPO, and POD activities**

PAL, PPO, and POD enzyme activities in the two different types of packages were significantly different (Fig. 3). PAL activity in the two different packaging samples was increased, and there was no significant difference during the early stage of storage (the first 2 weeks) (Fig. 3A). However, PAL enzyme activity increased rapidly, ranging from 8.708 U/g on day 0 to 12.225 U/g on day 35 in the control group; however, vacuum-packaged samples showed no significant change in PAL activity. The PPO activity in the vacuum packaging was lower than that in the atmospheric pressure packaging during the whole storage period. The initial PPO value ranged from 0.396 to 0.604 U/g in the vacuum group and 1.178 U/g in the atmospheric pressure group on day 35 (Fig. 3B). This indicated that PPO enzyme activity was very sensitive in vacuum packaging, and that the PPO enzyme is the key enzyme for the browning of fruits and vegetables; these results were consistent with those of a previous study (Min et al., 2017). Similar to PAL, the most significant difference in activity between packaging methods became apparent after day 14: POD activity in the vacuum group samples increased from 0.528 U/g on day 14 to 0.604 U/g on day 35, whereas activity in the atmospheric group increased from 0.904 U/g to 1.178 U/g during the same time period (Fig. 3C). Finally, the PAL, PPO, and POD enzymatic activities in the vacuum packaging were lower than those in the atmospheric pressure packaging, which indicated that the vacuum conditions could effectively inhibit enzyme activity and, thus, inhibit enzymatic browning (Xing et al., 2012).

During this experiment, the changes in PAL, PPO, and POD activities under different packaging conditions were studied, and the changes were consistent with the browning of fresh-cut lotus root during storage; these results were consistent with those of previous reports (Banerjee et al., 2015; Cheng et al., 2015).

**Gene expression of NnPAL, NnPPO, and NnPOD**

Two different types of packaging had different effects on the two NnPAL genes (Fig. 4). NnPAL1 expression in atmospheric packaging increased gradually during the whole storage period, reaching a peak on day 28. NnPAL1 was continuously repressed by vacuum packaging, and the transcript abundance was decreased by more than 60-fold on day 28. NnPAL2 mRNA accumulation increased in vacuum packaging, and the levels were relatively lower than those for NnPAL1. It was suggested that the expression of NnPAL1 mRNA paralleled the changes in PAL enzyme activity and browning degree, and that NnPAL1 was most likely to be involved in the synthesis of phenolic compounds in lotus root browning, which was supported by our previous work (Min et al., 2017). NnPPOA transcripts were increased in atmospheric pressure packaging, whereas it remained constant in vacuum packaging, which suggested that vacuum packaging inhibited NnPPOA expression. Similar to NnPPOA, NnPODAC transcripts were increased...
in atmospheric pressure packaging, whereas it decreased on day 1 and remained constant in vacuum packaging, but with much lower levels (Fig. 5). It was suggested that NnPPOA mRNA changes were consistent with the PPO enzyme activity and browning degree. Furthermore, NnPPOA was most likely involved in lotus root browning. Similar results have been reported by other studies (Min et al., 2017).

In the NnPOD gene family, NnPOD2/3 mRNA were significantly decreased in vacuum packaging compared with atmospheric packaging during the whole storage time (Fig. 6). This phenomenon indicated that vacuum packaging inhibited the expression of these two genes, which was consistent with the changes in POD enzyme activity and browning degree. NnPPOD1/5 were inhibited during early storage and upregulated during late storage in vacuum packaging. NnPPOD4/6 were induced by vacuum packaging during the whole storage period. Nevertheless, there was no significant difference in NnPPOD7 expression between the two packaging methods. It was suggested that the expression patterns of NnPPOD2/3 were consistent with the changes in POD enzyme activity and browning degree; therefore, they may be key genes involved in lotus root browning.

Our previous work found that downregulation of NnPAL1, NnPPO4, and NnPPO2–4 was consistent with the decreased browning of fresh lotus roots at low temperatures for two different varieties (‘E lian 6’ and ‘E lian 5’) (Min et al., 2017). Changes in NnPAL1, NnPPO4, and NnPPO2/3 expressions occurred in parallel with changes in PAL, PPO, and POD enzyme activities and the browning degree in vacuum and atmospheric packaging. Based on the present results and our previous report, NnPAL1, NnPPO4, and NnPPO2/3 are the key genes affecting the browning of fresh-cut lotus root; however, further functional analyses should be performed to confirm these results.

**ERF gene expression**

Seven ERF genes were isolated during our previous study. These ERF genes showed diverse expression patterns (Fig. 7). NnERF2 was induced by vacuum packaging during the early stage; however, it was induced by atmospheric pressure packaging during the later stage. There was no obvious difference in the expression of NnERF3 in the two types of packaging. NnERF4 expression increased and then decreased during storage in the two types of packaging, but it was always lower in vacuum packaging than in atmospheric pressure packaging during the early and middle storage periods (days 7, 14, and 21), although the expression was relatively low. NnERF5 mRNA increased gradually during the whole storage period, peaking on day 35, but NnERF5 was significantly upregulated in vacuum packaging compared with that in atmospheric packaging. NnERF1/6/7 were significantly induced by vacuum packaging, and the transcript abundance was increased by more than 800-, 400-, 700-fold, respectively.

NnPPOA expressions were significantly suppressed by vacuum packaging, which was consistent with NnPAL1, NnPPO4, and NnPPO2/3 changes and browning. This indicated that in lotus root, the decreased expressions of NnPPO4/5 were concurrent with decreases in browning; however, the specific relationship between ERF and browning is unclear and should be studied further. In our previous study, we found that NnPPO3/4/5 were continuously downregulated by low temperatures and were associated with the browning of fresh-cut lotus roots under low-temperature conditions. Therefore, it was proposed that NnPPO3/4/5 could be important regulators of fresh-cut

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Fig. 4. Messenger RNA (mRNA) from phenylalanine ammonia lyase (PAL) genes in response to different temperature treatments. Fresh-cut lotus root was separately stored in vacuum packaging (VP) (black) and atmospheric packing (AP) (red) at 4 ºC. The day 0 sample values were set at 1. Error bars represent SEs from three biological replicates.

Fig. 5. Messenger RNA (mRNA) from polyphenol oxidase (PPO) genes in response to different temperature treatments. Fresh-cut lotus root was separately stored in vacuum packaging (VP) (black) and atmospheric packing (AP) (red) at 4 ºC. Error bars represent SEs from three biological replicates.

Fig. 6. Messenger RNA (mRNA) from peroxidase (POD) genes in response to different temperature treatments. Fresh-cut lotus root was separately stored in vacuum packaging (VP) (black) and atmospheric packing (AP) (red) at 4 ºC. Error bars represent SEs from three biological replicates.
lotus root browning. We further studied the effects of vacuum packaging on browning and ERF gene expression in lotus root. NnERF4/5 were highly correlated with the browning process of lotus root, which further supported the previous findings. Therefore, NnERF4/5 were proposed to be important regulators of fresh-cut lotus root browning. Moreover, because of the significantly different expression levels of ERF4 and ERF5 during storage, and because of the differences in the response to vacuum packaging compared with ERF4, it seemed that NnERF5 was more positively correlated with browning, and that ERF4/5 might have different roles in lotus root browning. ERF4 mainly participated in lotus root browning during early storage, whereas ERF5 functioned during the whole storage period. The relationships of NnERF4/5 and NnPPOA, NnPPOA, and NnPPO2/3 during lotus root browning should be the subject of further research. NnERF1/6/7 were significantly increased during vacuum storage, which indicated that they are more sensitive to hypoxia stress (vacuum storage). The processing of fresh-cut fruits and vegetables promotes physiological and biochemical changes in the product (Toivonen and Brummell, 2008). These changes may cause significant decreases in color, texture, and flavor. Fresh-cut peppers in vacuum packaging had more noticeable ethanol and acetaldehyde contents, which are essential to the flavor of fresh-cut products (González-Aguilar et al., 2004). Therefore, NnERF1/6/7 may be related to the loss of quality of fruits and vegetables, especially the loss of flavor. Similar studies involving two hypoxia-responsive ERF (DkERF9 and DkERF10) genes showed acetaldehyde and ethanol synthesis in persimmon were separately regulated by DkPDC and DkADH promoters (Min et al., 2014).

Conclusions

Browning and PAL, PPO, and POD enzyme activity and gene expression results of this study showed that vacuum packaging effectively delayed lotus root browning. The downregulation of NnPPOA, NnPPOA, and NnPPO2/3 in vacuum packaging coincided with the increased related enzyme activities and the browning degree of fresh-cut lotus root. Furthermore, the expression patterns of NnERF4/5 were consistent with the changes in NnPPOA, NnPPOA, and NnPPO2/3 gene expressions. It has been proposed that NnERF4/5 could be important regulators of fresh-cut lotus root browning.

Literature Cited

Abraham, M., J. Zouine, and I.E. Hadrami. 2011. Low concentrations of BAP and high rate of subcultures improve the establishment and multiplication of somatic embryos in date palm suspension cultures by limiting oxidative browning associated with high levels of total phenols and peroxidase activities. Scientia Hort. 130:344–348.

Banerjee, A., P. Suprasanna, S. Vvariyan, and A. Sharma. 2015. Gamma irradiation inhibits wound induced browning in shredded cabbage. Food Chem. 173:38–44.

Baxter, J.H. 2010. Free Amino Acid Stability in Reducing Sugar Systems. J. Food Sci. 60:405–408.
and volatile composition of mulberry wine. J I Brewing. 124:45–56.
Lu, S., Y. Luo, E. Turner, and H. Feng. 2007. Efficacy of sodium chlorite as an inhibitor of enzymatic browning in apple slices. Food Chem. 104:824–829.
Müller, M. and S. Munne-Bosch. 2015. Ethylene Response Factors: A Key Regulatory Hub in Hormone and Stress Signaling. Plant Physiol. 169:32–41.
Mcdonald, K. and D.W. Sun. 2000. Vacuum cooling technology for the food processing industry: A review. J Food Eng. 45:55–65.
Min, T., F. Fang, H. Ge, Y.N. Shi, Z.R. Luo, Y.C. Yao, D. Grierson, X.R. Yin, and K.S. Chen. 2014. Two novel anoxia-induced ethylene response factors that interact with promoters of deastringency-related genes from persimmon. PLoS One 9:97043.
Min, T., M.M. Wang, H.X. Wang, X.F. Liu, F. Fang, D. Grierson, X.R. Yin, and K.S. Chen. 2015. Isolation and expression of NAC genes during persimmon fruit postharvest astringency removal. Intl. J. Mol. Sci. 16:1894–1906.
Min, T., J. Xie, M.L. Zheng, Y. Yi, W.F. Hou, L.M. Wang, Y.W. Ai, and H.X. Wang. 2017. The effect of different temperatures on browning incidence and phenol compound metabolism in fresh-cut lotus (Nelumbo nucifera G.) root. Postharvest Biol. Technol. 123:69–76.
Min, T., X.R. Yin, Y.N. Shi, Z.R. Luo, Y.C. Yao, D. Grierson, I.B. Ferguson, and K.S. Chen. 2012. Ethylene-responsive transcription factors interact with promoters of ADH and PDC involved in persimmon ( Diospyros kaki ) fruit de-astringency. J. Expt. Bot. 63:6393–6405.
Nakano, T., K. Suzuki, T. Fujimura, and H. Shinshi. 2006. Genome-wide analysis of the ERF gene family in Arabidopsis and rice. Plant Physiol. 140:411.
Phukan, U.J., G.S. Jeena, V. Tripathi, and R.K. Shukla. 2017. Regulation of Apetala2/Ethylene response factors in plants. Front. Plant Sci. 8:150.
Pma, T. 2006. Fresh-cut apples: Challenges and opportunities for multi-disciplinary research. Can. J. Plant Sci. 86:1361–1368.
Qin, L., L. Wang, Y. Guo, Y. Li, H. Umit, and Y. Wang. 2017. An ERF transcription factor from Tamarix hispida, ThCRF1, can adjust osmotic potential and reactive oxygen species scavenging capability to improve salt tolerance. Plant Sci. 265:154.
Shan, L.I., Y. Zhu, F.U. Da-Qi, B.Z. Zhu, R. Hao, and Y.B. Luo. 2013. Effect of chitosan coating combined with oxygen-free packaging on enzymatic browning during cold storage of young lotus rhizomes. Food Sci. 34:243–251.
Soliva-Fortuny, R.C. and O. Martín-Belloso. 2003. New advances in extending the shelf-life of fresh-cut fruits: A review. Trends Food Sci. Technol. 14:341–353.
Son, J., J.E. Hyun, J.W. Lee, S.Y. Lee, and B. Moon. 2015. Combined application of antibrowning, heat treatment and modified-atmosphere packaging to extend the shelf life of fresh-cut lotus root. J. Food Sci. 80:C1178–C1187.
Tan, Y.T. and K.F. Zeng. 2014. Effects of combined treatment with ascorbic acid, cysteine and CaCl2 on browning of fresh-cut taro. Food Science 35:231–235.
Toivonen, P.M.A. and D.A. Brummell. 2008. Biochemical bases of appearance and texture changes in fresh-cut fruit and vegetables. Postharvest Biol. Technol. 48:1–14.
Tsouvatzis, P., A. Deltisidis, and J.K. Brecht. 2011. Hot water treatment and pre-processing storage reduce browning development in fresh-cut potato slices. HortScience 46:1282–1286.
Xing, Y., X. Li, Q. Xu, J. Yun, and Y. Lu. 2012. Extending the shelf life of fresh-cut lotus root with antibrowning agents, cinnamon oil fumigation and moderate vacuum packaging. J. Food Process Eng. 35:505–521.
Yang, C.Y., F.C. Hsu, J.P. Li, N.N. Wang, and M.C. Shih. 2011. The AP2/ERF transcription factor AtERF73/HRE1 modulates ethylene responses during hypoxia in Arabidopsis. Plant Physiol. 156:202–212.
Yang, R., J. Liu, Z. Lin, W. Sun, Z. Wu, H. Hu, and Y. Zhang. 2018. ERF transcription factors involved in salt response in tomato. Plant Growth Regulat. 84:573–582.
Zhou, Y., J.M. Dahler, U. Sjr, and W. Rhb. 2003. Enzymes associated with blackheart development in pineapple fruit. Food Chem. 80:565–572.