Chlorophyll Degradation and Lignification of Fresh-cut Water Fennel Treated with a Complex Chemical Solution and Subsequent Packaging

Runqiang Yang, Jingru Wang, Xintian Nie¹, Yan Zhuang, Zhenxin Gu*, and Qianghui Guo

College of Food Science and Technology, Nanjing Agricultural University, Nanjing 210095, China

¹College of Engineering, Nanjing Agricultural University, Nanjing 210000, China

Received September 8, 2015
Revised December 22, 2015
Accepted December 28, 2015
Published online April 30, 2016

*Corresponding Author
Tel/Fax: +86-025-84396293
E-mail: guzx@njau.edu.cn

pISSN 1226-7708
eISSN 2092-6456

© KoSFoST and Springer 2016

Abstract Fresh-cut water fennel was treated with a novel complex chemical solution and subsequently packaged prior to refrigerated storage. Chlorophyll degradation and lignification were investigated. The complex solution consisted of 1.16 g/L calcium lactate, 150 mg/L zinc acetate, and 15.29 mg/L salicylic acid. Chlorophyllase, Mg-dechelataase, and chlorophyll-degrading peroxidase activities of water fennel decreased after immersion in the complex solution, which delayed chlorophyll degradation and increased L^* and a^* values. Complex solution treatment also reduced phenylalanine ammonia-lyase (PAL) activity and, thus, inhibited accumulation of lignin. After the complex solution treatment, packaging in high-density polypropylene plastic bags further retained chlorophylls and alleviated lignification of water fennel. Treatment with a complex chemical solution and subsequent packaging can maintain the quality, delay senescence, and improve the shelf life of water fennel.

Keywords: *Oenanthe javanica*, fresh-cut, packaging, chlorophyll degradation

Introduction

Water fennel (*Oenanthe javanica* D.C), locally called water dropwort or water fennel, is a biennial aquatic plant of the Apiaceae family that is widely cultivated in the Yangtze River area of China, and in east Asia in Japan, India, Vietnam, and Korea. Recently, water fennel was also introduced into Italy and other European countries (1). The edible part of water fennel is the stem, which is rich in chlorophyll, crude protein, vitamins, and dietary fiber, and minerals, including iron, calcium, and phosphorus (2). Dietary consumption of water fennel can reduce blood pressure, enhance body immunity, and prevent cardiovascular diseases (3,4).

Fresh-cut vegetables, also known as minimally processed vegetables, can meet the needs of a modern fast-paced life due to freshness, healthfulness, and ready-to-cook features (5). In southern China, fresh cut water fennel has gradually appeared in city markets. However, commercially use of water fennel is greatly limited due to storage intolerance. After processing, water fennel has a high water content that maintains life activities. During storage and transportation, general deterioration of fresh-cut water fennel involves degradation of chlorophyll and formation of lignin (6). Consequently, color, flavor, texture, and health-promoting effects are negatively influenced. The process of chlorophyll degradation is closely associated with the

enzymes chlorophyllase, Mg-dechelataase, and chlorophyll-degrading peroxidase (7). Phenylalanine ammonia-lyase (PAL) is a key enzyme related to the first step of lignin biosynthesis (8,9). Therefore, maintenance of a green color and reduction of lignification can be achieved via inhibition of the activities of enzymes in fresh-cut water fennel during storage.

Several techniques have been used for retardation of chlorophyll degradation in vegetables during storage, including 1-methylcyclopropene application on parsley leaves (10), UV-C irradiation of Chinese Kale (11), and 6-benzylaminopurine (6-BA) (12) and heat treatments of broccoli florets (13). These treatments all inhibit enzyme activities involved in chlorophyll degradation. Ozone solution dipping (14), a calcium lactate treatment (15), and 6-BA can reportedly effectively inhibit lignification of fresh-cut green asparagus, lettuce, and *Zizania latifolia*, respectively. However, up to now, no additional methods have been applied for inhibition of chlorophyll degradation of fresh-cut water fennel.

Salicylic acid is reportedly linked to oxidative stress responses, which can inhibit lignification of many fruits (16). Nevertheless, whether salicylic acid acts as a component of complex solutions or as a single component solution when used alone, salicylic acid is rarely used for fresh maintenance of vegetables. Calcium lactate can inhibit lignification and a complex solution containing zinc acetate

reportedly extended the shelf life of Chinese jujube fruits (17). Packaging is a convenient and effective approach for retaining the quality of products like meat, fish, fruits, and vegetables (18). Zhu *et al.* (19) observed that packaging significantly inhibited chlorophyll degradation in spinach and celery, perhaps due to reduction in respiration caused by packaging. In this study, treatment of fresh-cut water fennel with a novel complex chemical solution consisting of calcium lactate, zinc acetate, and salicylic acid was used for an investigation of effects on chlorophyll degradation and lignification. The effect of packaging together with the complex solution was also evaluated.

Materials and Methods

Plant material Fresh water fennel was harvested at the mature stage from farmland in Nanjing, China in October of 2014 and immediately transported to the laboratory of College of Food Science and Technology, Nanjing Agricultural University. Plants of uniform size and color with no visual defects were selected and cooled with ice water for 20 min to reach a central temperature of $2\pm0.5^{\circ}\text{C}$ after rinsing with precooling water. Vegetables air-dried with a blowing machine (FH6251; FLYCO, Shanghai, China) were then cut into slices of 0.61 ± 0.12 cm in diameter and 15.5 ± 1.5 cm in length using a sharp stainless steel knife.

Complex chemical solution treatment Vegetable slices were immediately immersed in a complex solution containing 1.16 g/L calcium lactate, 150 mg/L zinc acetate, and 15.29 mg/L salicylic acid for 30 min at a 1:5 ratio of g of water fennel to mL of solution. This mixed solution was optimized using response surface methodology (RSM) in which the chlorophyll content retained 86.5% (data not shown). Then, immersed water fennel slices were divided into 2 equal groups. One group of 300 g was packaged in 45×35 cm, 6 μm thick high-density polypropylene plastic bags with O_2 and CO_2 permeability values of 40–50 and 23–25 $\text{mL}/(\text{m}^2\cdot24\text{ h})$, respectively, sealed, and labeled as “Treated+Packed”. The other unpackaged group was labeled as “Treated+Unpacked”. Control samples were treated with distilled water, not packaged, labeled as “Control”. The relative humidity of treatment without packaging was $63\pm2\%$. Subsequently, 10 groups per treatment were stored under refrigeration (Haier refrigerator, BCD-206STPA; Haier group, Shanghai, China) at 4°C for 20 days. Each treatment was replicated 3 times.

Color measurement Superficial vegetable color was evaluated using a Model CR-400 colorimeter (Minolta, Osaka, Japan) based on measurement of L^* and a^* values at 6 stem positions for each treatment, directly.

Lignin content determination Lignin amounts were gravimetrically determined following the methods of Luo *et al.* (8) and results were

expressed as percentages.

PAL activity assay PAL activity trials were conducted as described previously (8). One unit (U) of PAL activity was defined as the amount of enzyme that resulted in an increase in absorbance of 0.01 at 290 nm in 1 h under test conditions (UV/vis spectrophotometer, UV-2802; UNICO, Dayton, NJ, USA).

Determination of total chlorophyll, chlorophyll a and b contents

Chlorophyll concentrations were determined following Lichtenthaler (20) with modification. A 5 g water fennel was ground using a mortar and subjected to extraction using 25 mL of 95% ethanol, then was centrifuged at $10,000\times g$ for 15 min at 4°C (GL-20G-II; Shanghai Anting Scientific Instrument Factory, Shanghai, China). The supernatant was used for determination of the chlorophyll content at 665 and 649 nm (UV/vis spectrophotometer, UV-2802; UNICO), and the chlorophyll content was expressed as mg/g of fresh weight (FW).

Determination of chlorophyll-degrading enzyme activities

Chlorophyllase, Mg-dechelatase, and chlorophyll-degrading peroxidase were extracted following the methods of Fukasawa *et al.* (21) Acetone tissue powder (500 mg) was suspended in 8 mL of an 8 mmol/L phosphate buffer at pH 7.0 for chlorophyll-degrading peroxidase, or in 8 mL of an 8 mmol/L phosphate buffer at pH 7.0 containing 50 mmol/L KCl and 0.24% Triton X-100 for chlorophyllase and Mg-dechelatase. Crude enzyme extracts were stirred for 1.5 h at 4°C (Table concentrator, Changzhou Guohua Electric Appliance Co., Ltd., Changzhou, China), and centrifuged at $12,000\times g$ for 25 min at 4°C (GL-20G-II; Shanghai Anting Scientific Instrument Factory). Supernatants were collected for enzyme activity determinations.

Chlorophyllase activity: The chlorophyllase activity was determined as described by Amir-Shapira *et al.* (22) with modification. The reaction mixture consisted of 0.5 mL of an enzyme solution, 0.1 mL of 1.4% (v/v) Triton X-100, 0.2 mL of an 80 $\mu\text{g}/\text{mL}$ chlorophyll *a* acetone solution, and 0.5 mL of a 0.1 mol/L phosphate buffer at pH 7.0. The mixture was incubated (Incubator, Changzhou Guohua Electric Appliance Co., Ltd.) under darkness at 25°C for 30 min, then the reaction was stopped using addition of 4 mL of acetone. Remaining chlorophyll was extracted using 4 mL of hexane and assayed based on the absorbance value at 663 nm (UV/vis spectrophotometer, UV-2802; UNICO). Enzyme activity was expressed as an increment of optical density in the hydrophilic phase at 663 nm per min under specified conditions.

Mg-dechelatase activity: Mg-dechelatase activity determinations were carried out following a modified method of Suzuki and Shioi (23). The reaction solution contained 0.5 mL of a 50 mmol/L Tris-tricine buffer at pH 8.0, 0.3 mL of 100 μM chlorophyllin *a*, 0.4 mL of 1% Triton X-100, and 0.5 mL of crude enzyme solution. The activity was measured at 35°C based on reading a change in absorbance value at 686 nm. The Mg-dechelatase activity was expressed as an absorbance value increment at 686 nm per min under test conditions.

Chlorophyll-degrading peroxidase activity: The chlorophyll-degrading peroxidase activity was measured following a modification of the method reported by Yamauchi *et al.* (24). The reaction mixture contained a 100 mmol/L phosphate buffer at pH 5.5, 0.1% Triton X-100, a 20 µg/mL chlorophyll *a* acetone solution, 5.0 mmol/L *p*-coumaric acid, 0.3% hydrogen peroxide, and a crude enzyme solution in a total volume of 3.3 mL. The enzyme activity was assayed based on measurement of a decrease in absorbance value at 668 nm at 30°C.

Statistical analysis Data were expressed as a mean±standard deviation (SD) of 3 replications ($n=3$). SPSS 18.0 (SPSS Inc., Chicago, IL, USA) was used for statistical analysis. Duncan's multiple-range test was used at $p<0.05$ for significance.

Results and Discussion

Changes in fresh weight During storage, fresh weights of all water fennel decreased constantly (Fig. 1). The weight of complex solution-treated water fennel was significantly ($p<0.05$) higher than for the controls. Packaging also had a positive effect on the fresh weight as it retained the complex solution in treated and packaged water fennel.

Changes of surface color and total chlorophyll contents L^* and a^* values in control and treatment water fennel both increased during storage (Fig. 2A and 2B). However, values for complex solution-treated water fennel were significantly ($p<0.05$) lower than for controls. Packaged water fennel maintained significantly ($p<0.05$) lower L^* and a^* increases than unpackaged.

Total chlorophyll contents in all water fennel decreased continuously during storage (Fig. 2C). Complex solution treatments delayed loss of chlorophyll and retained significantly ($p<0.05$) higher chlorophyll contents than controls after storage for 5 days packaging with polypropylene plastic bags further retarded chlorophyll degradation in comparison with no packaging. At the end of storage, the chlorophyll content of complex solution treated-combined packaged water fennel was 57.1% of the initial value for fresh water fennel. Chlorophyll contents of complex solution-treated and unpackaged, and control water fennel, were 50.6 and 43.6%, respectively, of initial values.

During storage of post-harvest green plants, the most obvious characteristic of senescence is tissue yellowing caused by chlorophyll degradation, causing color change (11,25). In this study, yellowing of fresh-cut water fennel occurred after storage for 5 days, when the total chlorophyll content began to decrease significantly (Fig. 2C). The decrease in total chlorophyll content paralleled increases in L^* and a^* values with a significant negative correlation. Thus, the color of water fennel was closely related to the total chlorophyll content, consistent with a report of Chairat *et al.* (11), except for the difference that the H value was measured. However, Costa *et al.* (12)

Fig. 1. Weight changes in fresh-cut water fennel during storage. Different letters represent significant differences at $p<0.05$ for the same storage time.

reported that chlorophyll degradation occurred before visible color change in broccoli florets. Differences can be attributed to plant species, storage conditions, and seasons (25). Chlorophyll *a* and chlorophyll *b* contents both decreased continuously during storage, while the decrease rate of chlorophyll *a* was higher than for chlorophyll *b*, consistent with previous reports (12).

Changes in chlorophyll *a* and *b* contents Chlorophyll *a* contents in 3 water fennel all decreased with storage time (Fig. 3A). After treatment with the complex solution, the retention rate of chlorophyll *a* increased significantly ($p<0.05$), compared with controls. The content of chlorophyll *a* after 20 days storage was 60.6% of the initial value for complex solution-treated and packaged water fennel, 53.0% for complex solution treated without packaging, and 44.1% for controls. Packaging significantly ($p<0.05$) delayed loss of chlorophyll *a*, compared with controls.

A similar trend of decrease was also detected for chlorophyll *b* (Fig. 3B). The chlorophyll *b* content in complex solution treated water fennel was significantly ($p<0.05$) higher than for controls after the first 10 days of storage. There was no significant ($p>0.05$) difference during the late storage period after 15 and 20 days. Packaging presented no effects on chlorophyll *b* contents as packaged and unpackaged water fennel maintained the same level during the whole storage period with no significant ($p>0.05$) differences.

Chlorophyll degradation begins with removal of phytol to form chlorophyllides, which are still green, catalyzed by chlorophyllase (22). Then, elimination of Mg^{2+} from chlorophyllides is induced by Mg -dechelatase to form pheophorbide, where the green color is lost. Finally, pheophorbide is decomposed to fluorescent chlorophyll catabolites, which is colorless, via red chlorophyll catabolism by both Pheide *a* oxygenase and red chlorophyll catabolite reductase (7). Yamauchi *et al.* (26) suggested that chlorophyll-degrading peroxidase is also involved in chlorophyll degradation during the first step of chlorophyll *a* oxidation into 13²-hydroxychlorophyll *a*. Chlorophyll degradation in spinach leaves is mainly regulated by chlorophyll-

Fig. 2. Changes of L^* (A), a^* (B), and total chlorophyll content (C) of fresh-cut water fennel. Different letters represent significant differences at $p < 0.05$ for the same storage time.

degrading peroxidase (27). Herein, the total chlorophyll content (Fig. 2C) of chlorophylls a and b (Fig. 3) decreased constantly, perhaps related to chlorophyll enzymes, such as chlorophyllase, Mg-dechelatase, and chlorophyll-degrading peroxidase (7).

Changes in chlorophyllase, Mg-dechelatase, and chlorophyll-degrading peroxidase activities The chlorophyllase activity of control water fennel increased significantly ($p < 0.05$) during the first 15 days of storage, compared with controls, then decreased afterwards (Fig. 4A). The chlorophyllase activity of complex solution-treated without packaging water fennel increased slowly throughout the storage period but still significantly ($p < 0.05$) lower than for controls. After complex solution-treatment and subsequent

Fig. 3. Changes of chlorophyll a (A) and b (B) contents of fresh-cut water fennel. Different letters represent significant differences at $p < 0.05$ for the same storage time.

packaging, water fennel chlorophyllase exhibited a low activity that was significantly ($p < 0.05$) lower than for the other two groups.

The activity of Mg-dechelatase increased throughout senescence for both the control and treated water fennel (Fig. 4B). Similar to chlorophyllase, the activity of Mg-dechelatase in complex solution-treated water fennel was lower level than in controls during storage. Packaging further significantly ($p < 0.05$) inhibited an increase in Mg-dechelatase activity after stored for 10 days, compared with controls.

The chlorophyll-degrading peroxidase activity initially increased, then decreased during storage where a peak value was detected on the 15th day of storage (Fig. 4C). When water fennel was treated with the complex solution, the chlorophyll-degrading peroxidase activity was significantly ($p < 0.05$) lower than for controls. This kind of activity inhibition was further enhanced by packaging.

Activities of chlorophyllase, Mg-dechelatase, and chlorophyll-degrading peroxidase increased gradually during the first 15 days of storage (Fig. 4) and, thus, during the first 15 days, these 3 enzymes were all involved in chlorophyll degradation of water fennel. Chlorophyllase is considered to be latent with an activity that usually does not increase during senescence in most plants (7), and even declines (28). However, this study found that chlorophyllase activity in fresh-cut water fennel increased during storage. An increase in chlorophyllase activity was also previously reported in barley (29) and spinach (27), perhaps due to different plant species and organs

Fig. 4. Changes of chlorophyllase (A), Mg-dechelatase (B), and chlorophyll degrading peroxidase (C) activities of fresh-cut water fennel. Different letters represent significant differences at $p < 0.05$ for the same storage time.

studied. The Mg-dechelatase activity was negatively correlated with the total chlorophyll content in agreement with previous findings in broccoli florets (12). However, in Chinese Kale, the activity of Mg-dechelatase initially increased, then decreased (11). Thus, Mg-dechelatase plays different roles in chlorophyll degradation of different plant species.

Chlorophyll-degrading peroxidase possibly mediates chlorophyll degradation via catalyzing oxidation of a phenolic compound with the help of hydrogen peroxide, which finally degrades chlorophyll to a colorless compound (30). Hence, chlorophyll-degrading peroxidase is associated with lignification since lignification begins with oxidation of phenolics (9). In this study, there was a significant

Fig. 5. Changes in lignin content (A) and PAL activity (B) of fresh-cut water fennel. Different letters represent significant differences at $p < 0.05$ for the same storage time.

positive correlation between the water fennel lignin content and chlorophyll-degrading peroxidase activity for the first 15 days ($r = 0.986$, $p < 0.05$). The chlorophyll-degrading peroxidase activity increased during the yellowing of broccoli florets (13), and initially increased then subsequently decreased afterwards in Chinese Kale (11). Herein, for water fennel, the chlorophyll-degrading peroxidase activity exhibited a pattern of change similar to Chinese Kale, where a peak value was detected after 15 days of storage. These differences are related to plant species and to storage time. Broccoli florets and Chinese Kale were stored for 4 days and 8 days, respectively, while water fennel was stored for 20 days.

Changes of lignin content and PAL activity The lignin content of control water fennel increased during storage with an approximate 188.3% increase within 20 days (Fig. 5A). Complex solution treatment inhibited formation of lignin, while this inhibition effect was reinforced when water fennel was subsequently packaged. After storage for 20 days, the lignin content of complex solution treated and packaged samples was only 13.5%, whereas the unpackaged content was 21.0%.

The PAL activity exhibited the same trend as lignin with no significant ($p > 0.05$) difference between complex solution-treated and control groups during the first 5 days of storage (Fig. 5B). After storage for 10 days, the PAL activity of water fennel treated with the

complex solution was significantly ($p < 0.05$) lower than for controls. Besides, the increasing rate of PAL activity was effectively slowed by packaging. The PAL activity of complex solution-treated and subsequently packaged water fennel at 20 days of storage was just 25.3% of the control value.

Water fennel has high water content for maintenance of life activities during storage, resulting in lignification and lignin accumulation (Fig. 5A). The first step in lignin biosynthesis is conversion of phenylalanine to trans-cinnamic acid in the phenylpropanoid pathway catalyzed by PAL (8,9). Evidences has shown a close relationship between PAL activity and lignin biosynthesis (8). In this study, lignin accumulation paralleled an increasing PAL activity with a highly positive correlation ($r = 0.958$, $p < 0.05$). The novel complex solution decreased the PAL activity and the lignin content of water fennel (Fig. 5) attributed to salicylic acid in the complex solution. Salicylic acid is a simple phenolic compound that is widely present in plants that can activate plant anti-oxidant and anti-aging systems, resulting in inhibition of lignification (31). A salicylic acid treatment suppressed the PAL activity of loquat fruit and reduced the content of lignin (32).

In conclusion, complex chemical solution treatment and subsequent packaging delayed chlorophyll degradation of fresh-cut water fennel via inhibition of the activities of chlorophyllase, Mg-dechelate, and chlorophyll-degrading peroxidase. Treatments also effectively decreased the PAL activity and, thus, retarded accumulation of lignin. Besides, other nutritional qualities, such as crude protein, vitamins and dietary fiber contents, were retained. Therefore, a complex solution can be a useful method for maintaining post-harvest quality of fresh-cut water fennel during storage.

Acknowledgments Financial support was provided by the National Key Technology Support Program during the Twelfth Five-year Plan Period of China (2012BAD27B03), and the National Innovation Experimental Program for University Students (201510307059). This study was also a project funded by the Priority Academic Program Development of Jiangsu Higher Education Institutions (PAPD).

Disclosure The authors declare no conflict of interest.

References

- Baldini R, Maccioni S, Bedini G, Flamini G, Cioni P. Essential oils composition of *Melittis melissophyllum* L. and *Oenanthe pimpinelloides* L. (Liguria, Italy). Atti. Soc. Tosc. Sci. Nat. Mem. Serie. B 116: 61-66 (2009)
- Zhou X, Wang G. Nutrient concentration variations during *Oenanthe javanica* growth and decay in the ecological floating bed system. J. Environ. Sci. 22: 1710-1717 (2010)
- Park J-C, Yu Y-B, Lee J-H, Hattori M, Lee C-K, Choi J-W. Protective effect of *Oenanthe javanica* on the hepatic lipid peroxidation in bromobenzene-treated rats and its bioactive component. Planta Med. 62: 488-490 (1996)
- Zhou Y-f, Tang Q-y, Peng H, Wu L-z. Extraction and composition analysis of crude polysaccharide from young stems of *Oenanthe javanica*. Food Sci. 33: 138-142 (2012)
- Ahmed L, Martin-Diana AB, Rico D, Barry-Ryan C. Quality and nutritional status of fresh-cut tomato as affected by spraying of delactosed whey permeate compared to industrial washing treatment. Food Bioprocess Tech. 5: 3103-3114 (2012)
- Zhuang Y, Zhang T, Han Y, Gu Z. Effects of ice water precooling and storage temperature on physiological and biochemical characteristics of *Oenanthe javanica*. Food Sci. 34: 279-284 (2013)
- Matile P, Hörtensteiner S, Thomas H. Chlorophyll degradation. Annu. Rev. Plant Biol. 50: 67-95 (1999)
- Luo H, Jiang L, Zhang L, Jiang J, Yu Z. Quality changes of whole and fresh-cut *Zizania latifolia* during refrigerated (1 °C) storage. Food Bioprocess Tech. 5: 1411-1415 (2012)
- Humphreys JM, Chapple C. Rewriting the lignin roadmap. Curr. Opin. Plant Biol. 5: 224-229 (2002)
- Ella L, Zion A, Nehemia A, Amnon L. Effect of the ethylene action inhibitor 1-methylcyclopropene on parsley leaf senescence and ethylene biosynthesis. Postharvest Biol. Tech. 30: 67-74 (2003)
- Chairat B, Nutthachai P, Varit S. Effect of UV-C treatment on chlorophyll degradation, antioxidant enzyme activities and senescence in Chinese kale (*Brassica oleracea* var. *alboglabra*). Int. Food Res. J. 20: 623-628 (2013)
- Costa ML, Civello PM, Chaves AR, Martinez GA. Effect of ethephon and 6-benzylaminopurine on chlorophyll degrading enzymes and a peroxidase-linked chlorophyll bleaching during post-harvest senescence of broccoli (*Brassica oleracea* L.) at 20°C. Postharvest Biol. Tech. 35: 191-199 (2005)
- Funamoto Y, Yamauchi N, Shigenaga T, Shigyo M. Effects of heat treatment on chlorophyll degrading enzymes in stored broccoli (*Brassica oleracea* L.). Postharvest Biol. Tech. 24: 163-170 (2002)
- An J, Zhang M, Lu Q. Changes in some quality indexes in fresh-cut green asparagus pretreated with aqueous ozone and subsequent modified atmosphere packaging. J. Food Eng. 78: 340-344 (2007)
- Martin-Diana AB, Rico D, Frias J, Henehan G, Mulcahy J, Barat J, Barry-Ryan C. Effect of calcium lactate and heat-shock on texture in fresh-cut lettuce during storage. J. Food Eng. 77: 1069-1077 (2006)
- Asghari M, Aghdam MS. Impact of salicylic acid on post-harvest physiology of horticultural crops. Trends Food Sci. Tech. 21: 502-509 (2010)
- Wu H, Wang D, Shi J, Xue S and Gao M. Effect of the complex of zinc (II) and cerium (IV) with chitosan on the preservation quality and degradation of organophosphorus pesticides in Chinese jujube (*Zizyphus jujuba* Mill. cv. Dongzao). J. Agr. Food Chem. 58: 5757-5762 (2010)
- Sandhya. Modified atmosphere packaging of fresh produce: Current status and future needs. LWT-Food Sci. Technol. 43: 381-392 (2010)
- Zhu J, Xie J, Lin Y, Zhang H. Effect of temperature and packaging method on quality of two kinds of leafy vegetables. Food Machinery 4: 175-178 (2012)
- Lichtenthaler HK. Chlorophylls and carotenoids: Pigments of photosynthetic biomembranes. Method. Enzymol. 148: 350-382 (1987)
- Fukasawa A, Suzuki Y, Terai H, Yamauchi N. Effects of postharvest ethanol vapor treatment on activities and gene expression of chlorophyll catabolic enzymes in broccoli florets. Postharvest Biol. Tech. 55: 97-102 (2010)
- Amir-Shapira D, Goldschmidt EE, Altman A. Chlorophyll catabolism in senescing plant tissues: *In vivo* breakdown intermediates suggest different degradative pathways for citrus fruit and parsley leaves. P. Natl. Acad. Sci. 84: 1901-1905 (1987)
- Suzuki T, Shioi Y. Re-examination of Mg-dechelation reaction in the degradation of chlorophylls using chlorophyllin a as a substrate. Photosynth. Res. 74: 217-223 (2002)
- Yamauchi N, Harada K, Watada AE. *In vitro* chlorophyll degradation in stored broccoli (*Brassica oleracea* L. var. *italica* Plen.) florets. Postharvest Biol. Tech. 12: 239-245 (1997)
- Conte A, Conversa G, Scrocco C, Brescia I, Laverre J, Elia A, Del Nobile M. Influence of growing periods on the quality of baby spinach leaves at harvest and during storage as minimally processed produce. Postharvest Biol. Tech. 50: 190-196 (2008)
- Yamauchi N, Funamoto Y, Shigyo M. Peroxidase-mediated chlorophyll degradation in horticultural crops. Phytochem. Rev. 3: 221-228 (2004)
- Yamauchi N, Watada AE. Regulated chlorophyll degradation in spinach leaves during storage. J. Am. Soc. Hortic. Sci. 116: 58-62 (1991)
- Fang Z, Bouwkamp JC, Solomos T. Chlorophyllase activities and chlorophyll degradation during leaf senescence in non-yellowing mutant and wild type of *Phaseolus vulgaris* L. J. Exp. Bot. 49: 503-510 (1998)
- Sabater B, Rodruze MT. Control of chlorophyll degradation in detached leaves of barley and oat through effect of kinetin on chlorophyllase levels. Physiol. Plantarum 43: 274-276 (1978)
- Kato M, Shimizu S. Chlorophyll metabolism in higher plants VI. Involvement of peroxidase in chlorophyll degradation. Plant Cell Physiol. 26: 1291-1301 (1985)
- Dokhanieh AY, Aghdam MS, Fard JR, Hassanpour H. Postharvest salicylic acid treatment enhances antioxidant potential of cornelian cherry fruit. Sci. Hortic.-Amsterdam 154: 31-36 (2013)
- Wu J, Chen Q, Tang C, Xia H. Effects of exogenous salicylic acid on lignification and related enzymes activities of loquat during cold storage. T. Chinese Soc. Agr. Eng. 22: 175-179 (2006)