

Methyl Jasmonate Reduces Decay and Enhances Antioxidant Capacity in Chinese Bayberries

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The effects of methyl jasmonate (MeJA) on postharvest decay and antioxidant capacity in harvested Chinese bayberry fruit were investigated. Chinese bayberries were treated with 0, 1, 10, 100, or 1000 $\mu\text{mol L}^{-1}$ MeJA at 20 °C for 6 h and then stored at 0 °C for 12 days. MeJA at 10 $\mu\text{mol L}^{-1}$ was most effective in reducing fruit decay; quality parameters including pH value, total soluble solids, and titratable acidity were not significantly affected by MeJA treatment. Fruit treated with 10 $\mu\text{mol L}^{-1}$ MeJA exhibited significantly higher phenylalanine ammonia-lyase activity and higher levels of total phenolics, flavonoids, and anthocyanins as well as individual phenolic compounds than control. These fruits also maintained significantly higher antioxidant activity as measured by scavenging capacity against 1,1-diphenyl-2-picrylhydrazyl, superoxide, and hydroxyl radicals and by the reducing power test compared to the control. These results indicate that MeJA can effectively reduce fruit decay and improve antioxidant capacity of Chinese bayberry fruit.

KEYWORDS: Chinese bayberry; methyl jasmonate; decay; antioxidant capacity

INTRODUCTION

Chinese bayberry (*Myrica rubra* Seib & Zucc.), a subtropical fruit native to China, is popular for its flavor and attractive red to purple color. The fruit consists of capsule-like cellules termed flesh segments and is very susceptible to physiological deterioration and pathogen attack, limiting postharvest life to 1–2 days under ambient temperature (1). Chinese bayberry contains high levels of anthocyanins, flavonoids, and phenolic acids and is considered to be a good source of natural antioxidants, which may provide protection against various human diseases caused by oxidative stress. Previous study has shown that Chinese bayberry has high antioxidant activity against superoxide and hydroxyl radicals, and there is a positive correlation between antioxidant activity and total phenolic or anthocyanin content (2). Fruit maturity stage and postharvest storage conditions can also affect bioactive compound levels and antioxidant capacity in Chinese bayberry (3, 4).

Methyl jasmonate (MeJA), a naturally occurring compound, plays important roles in plant growth and development, fruit ripening, and responses to environmental stresses (5). It has been reported that MeJA treatment could effectively suppress postharvest diseases of various fruits including sweet cherry (6), loquat (7), peach (8), and grapefruit (9). In addition, it has been reported that a postharvest MeJA treatment maintained higher levels of bioactive compounds and enhanced antioxidant capacity in berry fruits including blackberries, raspberries, and strawberries (10–12). Because MeJA is already classified by the U.S. Food and Drug Administration as a Generally Recognized

As Safe (GRAS) substance, it may have potential commercial applications in postharvest treatments for quality maintenance by reducing decay and enhancing antioxidant activity. As antioxidant content is becoming an increasingly important parameter with respect to fruit and vegetable quality, it is of great interest to evaluate changes in antioxidant status during postharvest storage of horticultural crops. However, there are no published data on the effect of postharvest MeJA treatment on antioxidants and antioxidant activity in Chinese bayberry fruit.

The purpose of this study was to investigate the effect of MeJA treatment on fruit decay, total phenolics, total flavonoids, total anthocyanins, and antioxidant capacity as well as the main phenolic constituents in Chinese bayberry during storage at 0 °C.

MATERIALS AND METHODS

Chemicals. Commercial standards of gallic acid (purity \geq 99%), protocatechuic acid (purity \geq 97%), quercetin-3-*O*-rutinoside (purity \geq 97%), myricetin (purity \geq 96%), and cyanidin-3-glucoside (purity \geq 98%) were purchased from Sigma Chemical Co. (St. Louis, MO). Methanol, acetone, formic acid, and water were of HPLC grade and purchased from Scigene Co. (Nanjing, Jiangsu, China). 1,1-Diphenyl-2-picrylhydrazyl (DPPH), Folin–Ciocalteu, deoxyribose, nitroblue tetrazolium (NBT), riboflavin, methionine, ferric chloride, methionine, potassium ferricyanide, and trichloroacetic acid (TCA) reagents were of analytical grade and purchased from Sigma Chemical Co.

Fruit Materials and Treatment. Chinese bayberry (*M. rubra* Seib & Zucc. cv. Wumei) was hand-harvested at mature stage from an orchard in Suzhou, Jiangsu province, China, and transported within 4 h to our laboratory. Fruits were selected for uniform size, color, and absence of defects and then randomly divided into five lots. Each lot of fruit was placed in a 40-L airtight container for MeJA treatment. An appropriate amount of MeJA liquid (Aldrich Chemical Co., Milwaukee, WI) was

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spotted onto filter paper inside the containers and incubated at 20 °C for 6 h, allowing the MeJA to evaporate. MeJA concentrations of 0 (control), 1, 10, 100, or 1000 $\mu\text{mol L}^{-1}$ were calculated on the basis of the assumption that MeJA evaporated completely. Following treatment, the containers were opened, and all lots of fruit were stored at 0 °C and approximately 90% relative humidity for 12 days. There were three replicates of approximately 5 kg of fruit each per treatment. Fruit samples were taken before MeJA treatment (time 0) and at 3-day intervals during storage for decay evaluation and quality parameter analysis. Tissue samples from fruit without defects were mixed and frozen immediately in liquid nitrogen and then stored at -80 °C until used for analysis of phenylalanine ammonia-lyase activity (PAL), total phenolics, total flavonoid, total anthocyanin contents, and antioxidant capacity and for HPLC analysis for main individual phenolic constituents.

Fruit Decay. Fruit decay was visually estimated using 30 fruits from each replicate during the course of the experiment. Berries with visible mold growth were considered to be decayed. The severity of fruit decay was expressed as percent of fruit showing fungal symptoms.

Total Soluble Solids, Titratable Acidity, and pH Value. Ten fruits from each replicate were wrapped in cheesecloth and squeezed with a hand press, and the juice was analyzed for total soluble solids (TSS), titratable acidity (TA), and pH value. TSS was determined at 25 °C using a portable refractometer (WYT-4, Quanzhou, China). TA (as the percentage of citric acid) was determined by titrating 20 mL of bayberry juice to pH 8.2 with 0.1 mol L^{-1} NaOH. pH value was measured with a pH-meter (PHS-25B, Shanghai, China).

PAL Activity. Two grams of tissue sample was ground with a chilled mortar and pestle in 10 mL of 0.2 mol L^{-1} sodium borate buffer at pH 8.7 containing 20 mmol L^{-1} β -mercaptoethanol. The homogenate was then centrifuged at 20000g for 20 min (4 °C), and the supernatant was collected for PAL activity according to the method described by Cao et al. (7). One unit of PAL activity was defined as the amount of enzyme that caused an increase in absorbance of 0.01 at 290 nm in 1 h, and the results are expressed as units per milligram of protein.

Sample Preparation for Total Phenolics, Total Flavonoids, and Total Anthocyanin Contents and Antioxidant Capacity. To prepare the fruit extract, 5 g samples from each replicate were extracted twice with 10 mL of precooled 80% acetone containing 0.2% formic acid and then centrifuged at 18000g for 20 min (4 °C). The supernatant was combined and the final volume made to 25 mL for analysis of total phenolics, total flavonoids, total carotenoids, and antioxidant capacity.

Total Phenolics, Flavonoids, and Anthocyanins. Total phenolic content of bayberry extract was determined with Folin-Ciocalteu reagent according to the method of Slinkard and Singleton (13) using gallic acid as a standard. Results were expressed as milligrams of gallic acid equivalent (GAE) per 100 g of fresh weight.

Total anthocyanin content of bayberry extract was measured using the pH differential method (14). Absorbance was measured at 510 and 700 nm, respectively, in different buffers at pH 1.0 and 4.5, using $A = [(A_{510} - A_{700})_{\text{pH}1.0} - (A_{510} - A_{700})_{\text{pH}4.5}]$ with a molar extinction coefficient for cyanidin-3-glucoside of 29600. Results were expressed as milligrams of cyanidin-3-glucoside (C3G) equivalents per 100 g of fresh weight.

Total flavonoid content was measured according to method of Toor and Savage (15). Absorbance was measured at 510 nm, and results were expressed as milligrams of rutin equivalents per 100 g of fresh weight.

Scavenging Activities against Superoxide, Hydroxyl, and DPPH Radicals and Reducing Power. The assay of superoxide scavenging activity was based on the capacity of the extract to inhibit formazan formation by scavenging the superoxide radicals generated in a riboflavin-light-nitroblue tetrazolium system (16). The percentage inhibition of superoxide anion generation was calculated using the following formula: superoxide radical scavenging activity (%) = $100 - (\text{absorbance of sample/absorbance of control}) \times 100$.

Hydroxyl radical scavenging activity of the extract was determined according to the deoxyribose method described previously (17). The result was calculated according to the following formula: hydroxyl radical scavenging activity (%) = $100 - (\text{absorbance of sample/absorbance of control}) \times 100$.

The DPPH radical scavenging activity of the extract was estimated following the method of Larrauri et al. (18). The result was calculated

according to the following formula: DPPH radical scavenging activity (%) = $100 - (\text{absorbance of sample/absorbance of control}) \times 100$.

The reducing power of the extract was determined according to the method of Oyaizu (19). The result was expressed as the absorbance of mixtures measured at 700 nm.

HPLC Analysis of Anthocyanin and Phenolic Compounds. Individual phenolic compounds in Chinese bayberry tissue sample were separated and determined by high-performance liquid chromatography (HPLC) according to the method of Bao et al. (2) with some modifications. The supernatants from the extracts described above were concentrated to dryness using a rotavapor in a water bath at 35 °C, dissolved in 5 mL of acidified water (3% formic acid), and then passed through a C₁₈ Sep-Pak cartridge (Supelco Corp., Bellefonte, PA), which was previously activated with methanol followed by water and then 3% aqueous formic acid. Anthocyanins and other phenolics were adsorbed onto the column, whereas sugars, acids, and other water-soluble compounds were eluted with 10 mL of 3% formic acid. Anthocyanins and other phenolics were then recovered with 3.0 mL of acidified methanol containing 3% formic acid. The methanol extract was passed through a 0.45 μm membrane filter (Millipore Corp., Bedford, MA). HPLC analysis was carried out by means of an Agilent HPLC series 1100 equipped with 130 Chemstation software and a model G1315B diode array detector (Agilent Corp., Santa Clara, CA). Twenty microliter samples were injected at ambient temperature (20 °C) into a reserved-phase Nova-Pak C18 column (250 \times 5 mm, 5 μm , Agilent Corp.). The mobile phase consisted of 3% aqueous formic acid (A) and HPLC grade methanol (B) with a linear gradient from 0 to 14% B at the first 1 min, followed by a linear gradient from 14 to 60% B for the next 39 min, and then followed by 100% B for 5 min before returning to the initial condition. The flow rate was 1 mL min^{-1} , and the wavelengths of detection were set at 280, 370, and 520 nm. Scanning between 240 and 550 nm was performed. Retention times and spectra were compared to those of authentication standards.

Statistical Analysis. Experiments were performed using a completely randomized design. All statistical analyses were performed using SPSS 13.0 (SPSS Inc., Chicago, IL). The data were analyzed by two-way analysis of variance (ANOVA) with treatment and storage time as factors. The means were separated by Tukey's test, and differences at $p \leq 0.05$ were considered to be significant.

RESULTS

Fruit Decay and Quality. The effect of MeJA treatment on fruit decay significant ($p \leq 0.05$) varied with the concentrations applied and storage time. As shown in **Table 1**, treatment with 1 or 10 $\mu\text{mol L}^{-1}$ MeJA significantly ($p \leq 0.05$) inhibited fruit decay throughout the storage period, whereas high concentrations of MeJA (100 or 1000 $\mu\text{mol L}^{-1}$) inhibited fruit decay only for the first 6 days of storage and thereafter had little effect. MeJA at 10 $\mu\text{mol L}^{-1}$ was the most effective in controlling fruit decay among all of the treatments. At the end of the storage, decay incidence of 10 $\mu\text{mol L}^{-1}$ MeJA-treated fruit was only 10.12%, whereas 37.94% of fruit decay was shown in control fruit. TSS and TA contents of Chinese bayberry decreased gradually during storage. No significant ($p \geq 0.05$) differences in TSS and TA levels were observed among all of the treatments during 12 days of storage at 0 °C (**Table 1**). The pH value of bayberry fruit juice increased slightly, corresponding to a decrease in TA during storage. Little difference in pH value was observed among all of the treatments (**Table 1**).

PAL Activity and Total Phenolic, Anthocyanin, and Flavonoid Contents. MeJA at 10 $\mu\text{mol L}^{-1}$ treatment and storage time significantly ($p \leq 0.05$) affected PAL activity and total phenolic, anthocyanin, and flavonoid contents of Chinese bayberry (**Table 2**). As shown in **Figure 1A**, there was no notable change in PAL activity of the control fruit during storage at 1 °C for 12 days, whereas MeJA treatment markedly induced the increase of PAL activity and maintained significantly ($p \leq 0.05$) higher PAL activity during the whole storage. At the end of storage, the activity of PAL in MeJA-treated fruit was 65.6% higher than that

Table 1. Effects of MeJA Treatment on Decay Incidence, pH Value, and Total Soluble Solids (TSS) and Titratable Acidity (TA) Contents in Chinese Bayberry during Storage at 0 °C for 12 Days^a

| day | treatment | decay (%) | pH | TSS (%) | TA (%) |
|---------------------------|---------------------------|--------------|-------------|-------------|-------------|
| 0 | | 0.00 ± 0.00 | 3.04 ± 0.01 | 9.51 ± 0.06 | 0.80 ± 0.02 |
| 3 | control | 11.32 ± 0.17 | 3.10 ± 0.03 | 9.44 ± 0.11 | 0.76 ± 0.01 |
| | 1 μmol L ⁻¹ | 3.43 ± 0.14 | 3.07 ± 0.01 | 9.43 ± 0.07 | 0.77 ± 0.01 |
| | 10 μmol L ⁻¹ | 0.24 ± 0.01 | 3.11 ± 0.02 | 9.46 ± 0.03 | 0.78 ± 0.00 |
| | 100 μmol L ⁻¹ | 8.44 ± 0.26 | 3.09 ± 0.01 | 9.41 ± 0.06 | 0.74 ± 0.03 |
| | 1000 μmol L ⁻¹ | 9.45 ± 0.19 | 3.11 ± 0.02 | 9.47 ± 0.09 | 0.79 ± 0.05 |
| 6 | control | 21.62 ± 0.41 | 3.17 ± 0.03 | 9.21 ± 0.01 | 0.74 ± 0.03 |
| | 1 μmol L ⁻¹ | 11.83 ± 0.13 | 3.20 ± 0.02 | 9.24 ± 0.03 | 0.76 ± 0.05 |
| | 10 μmol L ⁻¹ | 3.32 ± 0.21 | 3.14 ± 0.02 | 9.22 ± 0.06 | 0.73 ± 0.05 |
| | 100 μmol L ⁻¹ | 17.51 ± 0.31 | 3.15 ± 0.01 | 9.17 ± 0.07 | 0.74 ± 0.02 |
| | 1000 μmol L ⁻¹ | 12.61 ± 0.58 | 3.16 ± 0.03 | 9.21 ± 0.03 | 0.77 ± 0.07 |
| 9 | control | 27.91 ± 0.31 | 3.21 ± 0.02 | 9.14 ± 0.04 | 0.74 ± 0.01 |
| | 1 μmol L ⁻¹ | 15.92 ± 0.21 | 3.19 ± 0.01 | 9.21 ± 0.04 | 0.71 ± 0.04 |
| | 10 μmol L ⁻¹ | 7.01 ± 0.1 | 3.22 ± 0.04 | 9.19 ± 0.12 | 0.76 ± 0.03 |
| | 100 μmol L ⁻¹ | 25.02 ± 0.43 | 3.20 ± 0.04 | 9.22 ± 0.05 | 0.74 ± 0.03 |
| | 1000 μmol L ⁻¹ | 31.17 ± 0.54 | 3.21 ± 0.00 | 9.21 ± 0.06 | 0.71 ± 0.03 |
| 12 | control | 37.94 ± 1.61 | 3.31 ± 0.02 | 9.00 ± 0.08 | 0.69 ± 0.01 |
| | 1 μmol L ⁻¹ | 21.88 ± 0.89 | 3.29 ± 0.01 | 8.97 ± 0.01 | 0.68 ± 0.02 |
| | 10 μmol L ⁻¹ | 10.12 ± 0.46 | 3.29 ± 0.02 | 9.04 ± 0.04 | 0.70 ± 0.02 |
| | 100 μmol L ⁻¹ | 37.43 ± 1.21 | 3.27 ± 0.01 | 9.04 ± 0.07 | 0.67 ± 0.03 |
| | 1000 μmol L ⁻¹ | 41.34 ± 1.51 | 3.28 ± 0.02 | 9.01 ± 0.01 | 0.69 ± 0.06 |
| significance ^b | | | | | |
| treatment (T) | df 3 | sig | ns | ns | ns |
| duration (D) | df 4 | sig | sig | sig | sig |
| T × D | df 12 | sig | ns | ns | ns |

^aData expressed as mean ± SE of triplicate assays. ^bns, not significant; sig, significant at $p \leq 0.05$.

Table 2. Two-Way ANOVA Outcomes for PAL Activity, Total Phenolics, Flavonoids, and Anthocyanins Contents, and Superoxide, Hydroxyl, and DPPH Radical Scavenging Capacities as well as Reducing Power of Chinese Bayberry Treated with 0 (Control) and 10 μmol L⁻¹ MeJA and Stored at 0 °C for 12 Days

| source | df | PAL activity | | total phenolics content | | total flavonoids content | | total anthocyanins content | | superoxide radical scavenging capacity | | hydroxyl radical scavenging capacity | | DPPH radical scavenging capacity | | reducing power | |
|---------------|----|--------------|------|-------------------------|------|--------------------------|-------|----------------------------|-------|--|------|--------------------------------------|-------|----------------------------------|------|----------------|------|
| | | F | P | F | P | F | P | F | P | F | P | F | P | F | P | F | P |
| treatment (T) | 1 | 243.85 | 0.00 | 158.96 | 0.00 | 34.54 | 0.00 | 73.21 | 0.00 | 109.14 | 0.00 | 44.51 | 0.00 | 148.19 | 0.00 | 285.82 | 0.00 |
| duration (D) | 4 | 59.80 | 0.00 | 84.79 | 0.00 | 15.63 | 0.00 | 16.20 | 0.00 | 501.56 | 0.00 | 55.68 | 0.00 | 64.40 | 0.00 | 171.30 | 0.00 |
| T × D | 4 | 49.83 | 0.00 | 23.16 | 0.00 | 2.98 | 0.045 | 5.43 | <0.01 | 9.37 | 0.00 | 2.91 | 0.048 | 23.77 | 0.00 | 35.88 | 0.00 |

in control. The total phenolic content in control fruit decreased gradually during storage, whereas total phenolic content in MeJA-treated fruit exhibited a slight increase over the first 3 days and, thereafter, decreased gradually during the last 9 days. MeJA treatment maintained significantly ($p \leq 0.05$) higher total phenolic content throughout the storage compared to the control (Figure 1B). The total anthocyanin content increased over the first 6 days and then decreased gradually during the remainder of storage. The levels of total anthocyanin were 28.9 and 22.1% higher in MeJA-treated fruit than in control on the 6th and 12th days, respectively (Figure 1C). The total flavonoid content decreased gradually during storage. MeJA treatment markedly inhibited the decrease in total flavonoid content and maintained a significantly ($p \leq 0.05$) higher level of total flavonoids than control fruit during storage (Figure 1D).

Antioxidant Activity. To evaluate the antioxidant activity of biological samples, several methods have been developed in recent years. We evaluated the antioxidant activity of Chinese bayberry fruit treated with 10 μmol L⁻¹ MeJA by the scavenging activity against superoxide, hydroxyl, and DPPH radicals and the reducing power test during the storage in this study. The influences of MeJA treatment and storage time on antioxidant activity were significant ($p \leq 0.05$) (Table 2). The superoxide radical scavenging activity in bayberry fruit decreased rapidly

with storage time. Fruit treated with MeJA tended to maintain significantly ($p \leq 0.05$) higher superoxide radical scavenging activity than control fruit during storage (Figure 2A). The hydroxyl radical scavenging activity in control fruit decreased gradually during storage, whereas in MeJA-treated fruit it exhibited a slight increase over the first 6 days followed by a gradual decrease during the remaining 6 days. Fruit treated with MeJA had a significantly ($p \leq 0.05$) higher hydroxyl radical scavenging activity during storage (Figure 2B). The DPPH radical scavenging activity and reducing power in control fruit declined with storage time, whereas they increased initially and reached a peak on the third day in MeJA-treated fruit. Significantly ($p \leq 0.05$) higher values of DPPH radical scavenging activity and reducing power were observed in MeJA-treated fruit compared to the control fruit during the whole storage (Figure 2C,D).

Anthocyanins and Phenolic Compounds. HPLC analysis of Chinese bayberry flavonoids revealed that several individual anthocyanins and phenolic compounds were presented in significant amounts (Table 3). Phenolic compounds such as gallic acid, protocatechuic acid, quercetin-3-*O*-rutinoside, myricetin, and cyanidin-3-glucoside, the major anthocyanin in Chinese bayberry, were detected. In general, all four phenolic compounds and cyanidin-3-glucoside increased gradually over the first 6 days and then decreased slightly during the remaining 6 days.

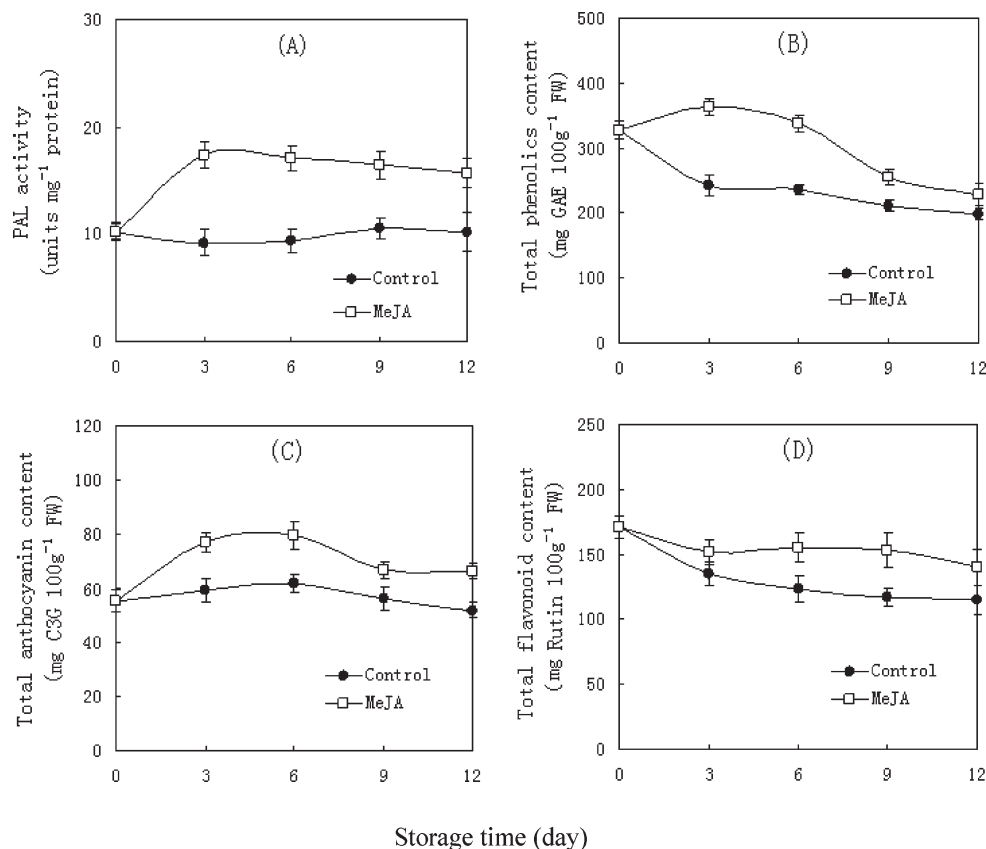


Figure 1. Effect of $10 \mu\text{mol L}^{-1}$ MeJA treatment on PAL activity (A) and total phenolic (B), anthocyanin (C), and flavonoid (D) contents of Chinese bayberry fruit during storage at 0 °C for 12 days. Data are expressed as mean \pm SE of triplicate assays. Vertical bars represent the standard errors of the means.

Significantly ($p \leq 0.05$) higher levels of protocatechuic acid, quercetin-3-*O*-rutinoside, myricetin, and cyanidin-3-glucoside were observed in MeJA-treated fruit during the whole storage time compared to the control fruit. Gallic acid contents were significantly ($p \leq 0.05$) higher in MeJA-treated fruit on the 9th and 12th days compared to the control fruit; however, no significant differences were found on the 3rd and 6th days of storage.

DISCUSSION

In the present study, we found that postharvest fungal decay of Chinese bayberry fruit was markedly affected by different MeJA concentrations. MeJA at $10 \mu\text{mol L}^{-1}$ was most effective in controlling fruit decay (Table 1). Similar results were also found in loquat (7) and grapefruit (9), with $10 \mu\text{mol L}^{-1}$ MeJA being the most effective concentration. In other studies, the effective concentration of MeJA on reducing fungal decay ranged from 1 to $1000 \mu\text{mol L}^{-1}$ (8, 20–22). These different results probably were due to the wide range in genetic makeup of different horticultural crops and different treatment methods of MeJA. Our results showed that 100 or $1000 \mu\text{mol L}^{-1}$ MeJA had little effect on fruit decay in bayberry fruit during the later storage period, which could be due to the enhanced fruit ripening and senescence by higher MeJA concentration. This dependency on concentration for MeJA efficacy has also been reported previously (23). Although MeJA has shown promise in preventing postharvest diseases in a number of horticultural crops, the mode of action of MeJA in reducing diseases is not well elucidated. It has been postulated that the control of postharvest diseases by MeJA is because of its direct inhibitory effect on pathogen growth and/or because of the induction of natural disease resistance (6, 7, 9, 23). It has been reported that natural disease resistance of fruits

is closely related to the degree of ripeness and senescence. The preformed natural antifungal compounds decreased during ripening with corresponding increase of disease incidence in some subtropical fruits (24). Moreover, cell wall disassembly and tissue softening during postharvest ripening render fruit more susceptible to pathogen infection and, hence, higher decay incidence (25). Therefore, it is possible that MeJA treatment might induce disease resistance or delay fruit ripening and senescence, thereby reducing decay incidence of Chinese bayberry fruit in this study. However, such action mechanisms need to be supported by further investigation.

As an endogenous phytohormone, MeJA plays key roles in regulating a great diversity of physiological and biochemical processes in plants including stimulating the biosynthesis of secondary metabolites (5). MeJA has been shown to induce stilbene accumulation in leaves and berries of grapevine plants (26), increase the accumulation of anthocyanins and phenolics in apples (27), raspberries (28), strawberries (29), and blackberries (12), and promote β -carotene synthesis in apple peel (30). PAL as a key enzyme in the first step of the phenylpropanoid pathway is directly involved in the biosynthesis of phenolic compounds, including phenols, stilbenes, and flavonoids (31). It has been reported that the accumulation of phenols and anthocyanins paralleled the increase in PAL activity in apple and grape fruits (32, 33). Therefore, the activity of PAL was examined in this study to investigate the possible role of PAL in phenolic metabolism of Chinese bayberry fruit in response to MeJA treatment. We found that fruit treated with $10 \mu\text{mol L}^{-1}$ MeJA exhibited significantly higher levels of PAL activity (Figure 1A), total phenolics, anthocyanins, and flavonoids (Figure 1B–D), and the main individual phenolic compounds (Table 3) compared to the control fruit. These results suggest that MeJA may improve the

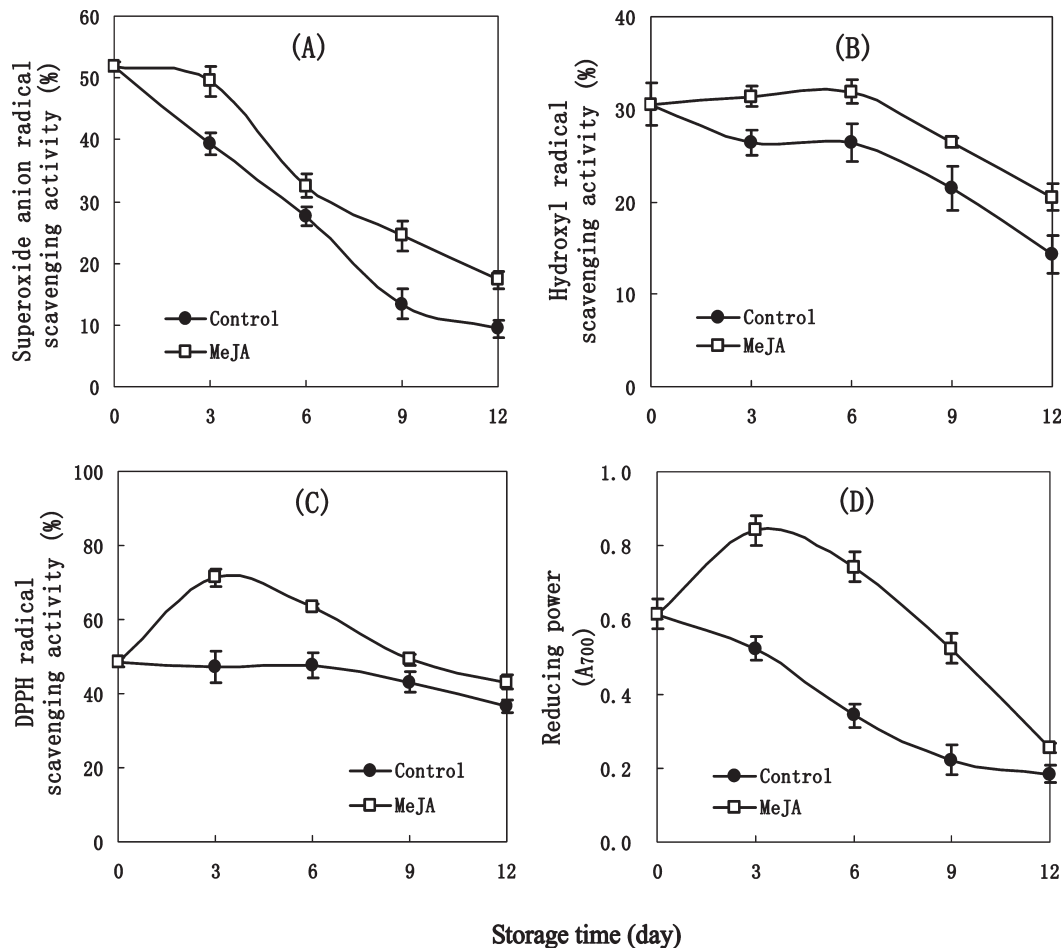


Figure 2. Effect of $10 \mu\text{mol L}^{-1}$ MeJA treatment on scavenging activities against superoxide anion (A), hydroxyl (B), and DPPH (C), radicals and reducing power (D) of Chinese bayberry fruit during storage at 0°C for 12 days. Data are expressed as mean \pm SE of triplicate assays. Vertical bars represent the standard errors of the means.

Table 3. Effect of $10 \mu\text{mol L}^{-1}$ MeJA Treatment on Gallic Acid, Protocatechuic Acid, Myricetin, Quercetin-3-O-rutinoside, and Cyanidin-3-glucoside Contents of Chinese Bayberry during Storage at 0°C for 12 Days^a

| day | treatment | gallic acid ^b | protocatechuic acid ^b | myricetin ^c | quercetin-3-O-rutinoside ^c | cyanidin-3-glucoside ^d |
|---------------------------|-----------|--------------------------|----------------------------------|------------------------|---------------------------------------|-----------------------------------|
| 0 | | 3.43 ± 0.54 | 1.80 ± 0.11 | 39.71 ± 1.54 | 17.29 ± 0.49 | 46.43 ± 2.15 |
| 3 | control | 4.11 ± 0.50 | 1.86 ± 0.33 | 43.43 ± 2.11 | 27.41 ± 1.11 | 47.66 ± 1.19 |
| | MeJA | 4.37 ± 0.26 | 2.73 ± 0.26 | 51.72 ± 1.39 | 31.60 ± 0.36 | 56.12 ± 1.49 |
| 6 | control | 4.55 ± 0.22 | 1.98 ± 0.14 | 52.30 ± 2.13 | 27.69 ± 1.12 | 53.83 ± 1.90 |
| | MeJA | 4.98 ± 0.13 | 3.17 ± 0.27 | 57.68 ± 0.95 | 32.62 ± 1.65 | 60.22 ± 2.06 |
| 9 | control | 3.99 ± 0.33 | 1.63 ± 0.35 | 45.89 ± 3.21 | 26.57 ± 0.80 | 45.11 ± 1.48 |
| | MeJA | 5.21 ± 0.71 | 2.68 ± 0.62 | 51.20 ± 1.53 | 31.46 ± 1.66 | 51.91 ± 1.73 |
| 12 | control | 3.53 ± 0.68 | 1.81 ± 0.21 | 39.78 ± 2.76 | 25.51 ± 1.43 | 42.25 ± 1.27 |
| | MeJA | 4.86 ± 0.91 | 2.90 ± 0.34 | 44.25 ± 1.68 | 30.62 ± 2.02 | 51.41 ± 2.38 |
| significance ^e | df | | | | | |
| treatment (T) | 1 | sig | sig | sig | sig | sig |
| duration (D) | 4 | sig | sig | sig | sig | sig |
| T \times D | 4 | sig | sig | sig | sig | sig |

^aData expressed as mean \pm SE of triplicate assays. ^bData expressed as micrograms per 100 g of fresh weight. ^cData expressed as micrograms of quercetin equivalents per 100 g of fresh weight. ^dData expressed as micrograms of cyanidin-3-glucoside equivalents per 100 g of fresh weight. ^esig, significant at $p \leq 0.05$.

antioxidant status of Chinese bayberry fruit by inducing PAL activity and thus positively affecting phenolic metabolism.

It has been reported that the antioxidant capacities of fruits and vegetables can be influenced by various pre- or post-harvest treatments. In several berry fruits including Chinese bayberry, the antioxidant activity was markedly enhanced by 60–100% oxygen treatments (4, 34, 35). MeJA has also been demonstrated

to be effective in enhancing antioxidant activity in berry fruits such as raspberries (11, 28), strawberries (10, 29), and blackberries (10, 12). In the present study, the scavenging activity against DPPH, superoxide, and hydroxyl radicals and the reducing power in MeJA-treated fruit were significantly higher than those in control fruit (Figure 2). This result suggests that a post-harvest application of MeJA will improve the health benefit

of Chinese bayberry fruit by enhancing the antioxidant activity. Previous research shows a significant positive relationship between total phenolic or anthocyanin content and antioxidant activity in some berry fruits (2, 34, 36, 37); thus, the higher radical scavenging activity and the reducing power in MeJA-treated bayberry fruit in the present study could be mainly attributed to its higher level of total phenolic or anthocyanin compounds.

In summary, a postharvest application of 10 $\mu\text{mol L}^{-1}$ MeJA maintained significantly higher levels of PAL activity, total phenolics, total anthocyanins, and total flavonoids as well as main individual phenolic and anthocyanin compounds than control fruit. In addition, MeJA markedly reduced fruit decay and maintained significantly higher antioxidant activity. Thus, MeJA has a potential application in postharvest treatment for reducing decay and maintaining a high-quality product in harvested Chinese bayberry fruit.

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