

Research Article

Chemical Composition and Antioxidant Activities of Five Samples of *Prunus mume* Umezu from Different Factories in South and East China

Zhi-Yi Chen, Yao-Sheng Lin, Xue-Ming Liu, Jing-Rong Cheng, and Chun-Ying Yang

Sericultural & Agri-Food Research Institute, Guangdong Academy of Agricultural Sciences, Key Laboratory of Functional Foods, Ministry of Agriculture, Guangdong Key Laboratory of Agricultural Products Processing, Guangzhou 510610, China

Correspondence should be addressed to Xue-Ming Liu; xuemingliu@21cn.com

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This paper investigated chemical composition and antioxidant activities of umezu, pickling liquid of *Prunus mume*, from different factories in South and East China. The organic acid and phenolic acid profiles were also analyzed. Results showed that umezu was rich in organic acids and extremely sour as *P. mume* fruit in addition to its high NaCl level ($\geq 20\%$). Total acid in umezu was more than 43.78 g/L in which main organic acids were citric acid and malic acid. Umezu contained more than 250.54 mg GAE/L total phenolic in which dominant phenolic acids were hydroxycinnamic acid derivatives. Umezu exhibited powerful antioxidant activities in ORAC, ABTS, DPPH, and FRAP assays. Reducing sugar, phenolic compounds, and antioxidant activities of umezu were affected by sample origins and fruit cultivars. Given its rich flavor components and high antioxidant activity, umezu could serve as a new dietary supplement or a natural preservative in food industry.

1. Introduction

Prunus mume, also known as Mei or green plum in China, mume or Japanese apricot in Japan, and Maesil in Korea, is a deciduous tree of the Rosaceae family [1]. The fruit of *P. mume*, weighing 12.0–30.0 g freshly, green in immature and yellow in mature stage as well as rich in organic acids, edible fibre, minerals, and phenolic compounds, is known to have good medicinal effects on stomach and intestine disorders [2]. Unlike many other fruits which are mainly consumed freshly, most *P. mume* fruits should be processed before consumption, due to presence of cyanoglycoside, prunasin, and amygdalin and very high organic acid content [3].

Traditionally, *P. mume* fruit was firstly pickled with salt or smoked to dry and then processed to preserved fruit or used as medicinal material [4]. For example, the pickled *P. mume* fruit (named umeboshi in Japan) has been widely used as a food garnish, sauce, juice, and liquor in China, Japan, Korea, and other Southeast Asian countries. Besides, smoked and dried immature *P. mume* fruit, *Fructus mume* (Chinese name “Wu Mei”), has been traditionally used as a medical

alternative for its antitussive, expectoration, antiemetic, antidiarrheal, anthelmintic, and antipyretic actions [5]. In addition, in many other Asian countries, mainly in Japan, a few of these processed products of *P. mume* fruit, such as umeboshi and *bainiku ekisu* (concentrated juice of *P. mume* fruit), had been used as a folk remedy for fatigue, diarrhea, and fever [6].

Even though a growing number of new technologies have been developed, salt-pickling remains the dominant method for *P. mume* fruit processing. During pickling of *P. mume* with salt, a large amount of liquid named Meilu in China and umezu in Japan is produced. Owing to its extreme salty and sour taste, umezu is mostly disposed as sewage, causing environment pollution. Umezu, composed of salt and liquid from *P. mume* fruit, is a natural source with great potential application. To realize the industrial utilization of umezu, its nutritional values, functional properties, and potential disadvantages should be characterized. To the best of our knowledge, there are no previous reports on umezu compositional characteristics, as well as further development. In this paper, five *P. mume* pickling liquid (umezu) samples

were collected from different factories in the South and East China, and their chemical constituents, including the content and proportion of organic acids and phenolic acids, and antioxidant activities were investigated.

2. Materials and Methods

2.1. Chemicals and Reagents. Folin-Ciocalteu phenol reagent, Trolox® (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid), 2,4,6-tripyridyl-s-triazine (TPTZ), 2,2'-azinobis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS), 2,2'-azobis-(2-amidinopropane) dihydrochloride (ABAP), and 3',6'-dihydroxyspiro[isobenzofuran-1(3H), 9'-(9H)-xanthene]-3-one disodium salt (FL) were purchased from Sigma Chemical Co. (St. Louis, MO, USA). Gallic, chlorogenic, neochlorogenic, cryptochlorogenic, caffeic, p-coumaric, protocatechuic, vanillic, and ferulic acids, catechin, and epicatechin were purchased from Biopurify Phytochemicals Ltd (Chengdu, China). HPLC-grade acetic acid and acetonitrile were obtained from Fisher (Suwanee, GA, USA). All other chemicals used were of analytical grade or above.

2.2. Umezu Samples. Two umezu samples (umezu of white mume and green mume) were acquired from Zhaoan (Fujian Province, China), while the other three samples were from Puning, Raoping, and Luhe (Guangdong Province, China), respectively, in May 2015. Both Baifen Mei and Qingzhu Mei were two dominating cultivars of *P. mume* planted in China. All umezu samples were stored in pots made of polyethylene terephthalate (PET) under 4°C until test.

2.3. Proximate Composition. Umezu samples were centrifuged at 3130 ×g for 10 min before test. A benchtop refractometer (RFM340, Bellingham and Stanley, UK) was used to acquire soluble solids content (SSC). The pH value was assayed with a pH meter (PB -20, Sartorius, Göttingen, Germany). The NaCl content was determined following the Chinese National Standard (GB/T 12457-2008). TA content was determined according to AOAC method 942.15 [7]. Aliquot of 2.0 mL sample was diluted to 100 mL with ultra-pure water, mixed, and then titrated with 0.1 M NaOH to an end point of pH 8.2. TA was calculated as % citric acid. The total phenolic content was analyzed by the Folin-Ciocalteu (FC) colorimetric method [8]. Briefly, 0.3 mL neutralized umezu was mixed with 3.7 mL of distilled water and 2.0 mL of FC reagent for 6 min. A 2.0 mL of 7% aqueous sodium carbonate solution was added to make up a total volume of 8.0 mL. The mixture was incubated for 90 min at 25°C. The absorbance was detected at 760 nm using a Shimadzu UV-1800 spectrometer (Shimadzu Inc., Kyoto, Japan) using gallic acid as the standard. Total phenolic content was expressed as mg gallic acid equivalents (GAE) per 1000 mL sample. All the measurements were done in triplicate, and the average values were used.

2.4. Analysis of Organic Acids by HPLC. Determination of organic acid profile of umezu was conducted by high performance liquid chromatography (HPLC) as previously

described [9]. An Agilent 1200 series HPLC system (Agilent Technologies, Palo Alto, California, USA) equipped with VWD detector was used to analyze organic acids. The system was run at a flow rate of 0.1 mL/min. The samples were analyzed on an Aminex HPX-87H ion exchange column (300 mm × 7.8 mm) protected with a Bio-Rad guard cartridge (30 mm × 4.6 mm) composed of the same stationary phase and the column (not the guard cartridge) was thermostated at 35°C. Mobile phase was 5.0 mM H₂SO₄ in dH₂O. ChemStation Rev. A.10.02 software (Agilent Technologies) was used for data acquisition, peak integration, and standard calibration. Umezu samples were diluted with mobile phase (1:50). The injection volume was 20 μL. Organic acids were detected with the VWD detector at 210 nm. Samples were quantified by comparing the retention times and peak areas with the known authentic standards. The results were expressed as g/L.

2.5. Analysis of Phenolic Compounds by HPLC. Determination of the phenolic compounds was conducted by HPLC as previously described [10]. All samples were analyzed on an Agilent 1260 HPLC system (Agilent Technologies, Palo Alto, California, USA) equipped with DAD detector and autosampler, using a 250 × 4.6 mm, 5 μm Agilent Zorbax SB-C18 column (Palo Alto, CA, USA). The conditions used were as follows: column temperature 30°C; injection volume 20 μL; total HPLC run time 50 min; mobile phase 0.4% aqueous solution acetic acid (solution A); and acetonitrile (solution B). The mobile phase was programmed as follows: 0–40 min, solution B 5–25%; 40–45 min, solution B 25–35%; 45–50 min, solution B 35–50%. The mobile phase was pumped at a constant flow rate of 1.0 mL/min. Detection was set at 280 nm. Before analysis, all samples were filtered through a 0.25 μm membrane filter (Millipore, Billerica, MA, USA). Samples were quantified by comparing the retention times and peak areas with authentic standards. The results were expressed as mg/L.

2.6. Antioxidant Activity Determined by ORAC Assay. The oxygen radical absorbance capacity (ORAC) assay was carried out according to a modified method from our laboratory (Zhang et al., 2010). The assay was performed using black-walled 96-well plates (Corning Scientific, Corning, NY, USA). The diluted umezu was prepared with 75 mM phosphate buffer (pH 7.4). The final reaction consisted of 20 μL of umezu or 20 μL of Trolox standard (range = 6.25–50 μM) and 200 μL of fluorescein (final concentration 0.96 μM). The mixture was incubated at 37°C for 10 min. Twenty μL of 119 mM ABAP was then added to each well using a multi-channel pipette. Fluorescence intensity was measured using a Fluoroskan Ascent FL plate-reader (Thermo Labsystems, Franklin, MA, USA) at wavelengths of 485 nm for excitation and 538 nm for emission for 35 cycles every 4.5 min. ORAC was expressed as μM Trolox equivalents per 1000 mL.

2.7. Antioxidant Activity Determined by ABTS Assay. The ABTS radical scavenging activity assay was carried out as previously described [11]. Briefly, ABTS was dissolved in distilled water to a concentration of 7 mM. The ABTS stock solution was then obtained by mixing 7 mM ABTS water

solution with 2.45 mM potassium persulfate solution and leaving it to stand in the dark at room temperature for 12–16 h before use. The ABTS radical solution was diluted with methanol and adjusted to an absorbance of 0.700 ± 0.020 at 734 nm. The assay was performed using 96-well microplates. 0.10 mL of the diluted umezu was added to 2 mL of the ABTS radical solution for use in each reaction. The absorbance was measured at 734 nm. The ABTS antioxidant activity was expressed as $\mu\text{g/L}$ Trolox equivalents per 1000 mL.

2.8. Antioxidant Activity Determined by FRAP Assay. The FRAP assay was carried out according to the method as previously described [12]. The working solution consisted of 25 mL 300 mM acetate buffer (5.1 g $\text{CH}_3\text{COONa}\cdot 3\text{H}_2\text{O}$ and 20 mL of CH_3COOH pH 3.6), 2.5 mL TPTZ solution (10 mM TPTZ in 40 mM HCl), and 2.5 mL 20 mM $\text{FeCl}_3\cdot 6\text{H}_2\text{O}$ solution which was incubated 10 min at 37°C before use. A 0.2 mL umezu was reacted with 2.8 mL of the working solution for 30 min in the dark at room temperature. Absorbance was detected at 593 nm using a Shimadzu UV-1800 spectrometer. Methanol was used as the control and the standard curve was established with Trolox. The FRAP antioxidant activity was expressed as mg Trolox equivalents (TE) per 1000 mL umezu of sample.

2.9. Antioxidant Activity Determined by DPPH Assay. The DPPH radical scavenging capacities of umezu were performed according to a previous report [13]. Two hundred μL of six serially diluted umezu or methanol (control) samples were added to 2.8 mL of methanolic solution of $70 \mu\text{M}$ DPPH. After shaking, the mixtures were placed in the dark at room temperature for 30 min. A spectrometer (Shimadzu UV-1800) was used to read their absorbance values at 515 nm. Antioxidant activity was calculated according to the following equation in which $I\%$ stands for inhibition percentage: $I\% = (\text{Abs}_{\text{control}} - \text{Abs}_{\text{sample}}) \times 100 / \text{Abs}_{\text{control}}$. Sample concentration at which 50% DPPH scavenging effect was acquired was used to express median effective concentration (EC50) value. EC50 was calculated by constructing the percentage of DPPH scavenging versus log (phenolic concentration expressed as the volume of umezu) curves (mg/mL).

2.10. Statistical Analysis. All the analyses were performed in triplicate and results were presented as mean \pm standard deviations of each sample. Different samples were analyzed with ANOVA followed by the SNK-q test to identify differences between values. A value of $p < 0.05$ was considered to be statistically different. All statistical analyses were performed using the SPSS statistical package version 13.0 (SPSS Inc., Chicago, IL, USA).

3. Results and Discussion

3.1. Proximate Composition. Traditional *P. mume* pickling process is practiced in a cubic pool as follows: one layer of fruit by one layer of salt until the pool is full; and then an inert cloth is used to cover the surface and a heavy item, usually stone, is put on the top. Salt is added at a dose of

250–300 kg salt per 1000 kg fresh *P. mume* fruit. It takes more than three months for the pickling process to occur. During pickling, water and soluble substances such as sugar, organic acid, mineral, and bioactive compounds effuse from the fruits while salt is intruded into the fruits until a balance is achieved at the end. The umezu yield is 45–50% of fresh fruits.

The chemical composition could directly reflect the physiochemical property of *P. mume* umezu [4]. Table 1 summarized the proximate composition of umezu samples from five different factories in two provinces in China. As expected, most umezu samples were high in salt and acid but low in sugar. They contain 33.00–34.50° Brix solid matter in which 20.36–22.01% is NaCl. Total titratable acid content is 4.55–5.44%, which makes them very acidic (pH 1.85–1.90). Reducing sugar content is only 5.18–8.56%. The ratio of sugar to titratable acid among those samples is only 1.14–1.57. Noteworthy, the solid and NaCl contents of umezu from different factories were not different indicating similar pickling processing including recipe was used. Among all samples, umezu from Luhe presented the lowest total acid (4.55%) and reducing sugar (5.18%) while that of other four samples had similar total acid and reducing sugar contents. As reported previously, the *P. mume* fruit was an important source of phenolic antioxidant and antimicrobial agent [14–16]. The total phenolic content ranged from 250.54 to 330.74 mg/L in this study. The different phenolic content among the five umezu samples might be ascribed to different fruit cultivars and/or growing conditions as fruit quality was affected by factors including weather, environment, plant management, and so on besides species [17]. Thus, *P. mume* fruit from different species or different growing origins would render umezu samples with different chemical compositions.

3.2. Organic Acid Profile. Organic acids are important flavor regulators for acidity regulation that could confer sour flavor, control microorganism growth, prevent browning, increase solubility of metal ion, improve appetite, promote digestion, and so on [6]. The characteristic sourness of *P. mume* fruits is caused by the presence of free organic acids in this fruit [9, 18]. Generally, the total titratable acid of *P. mume* fruits could be as high as 6.0–7.0% on a fresh weight basis, though its profile is diversified [2]. Organic acids, with high polarity, are hydrophilic compounds and very stable in solution [19]. Chen et al. [18] detected seven organic acids in *P. mume* by HPLC. Pan (2007) purified ten organic acids, including citric acid, malic acid, oxalic acid, glycolic acid, succinic acid, lactic acid, pyroglutamic acid, formic acid, acetic acid, and propionic acid from this fruit. During pickling of *P. mume* fruit with salt, they could easily penetrate the fruit into the pickling liquids, that is, umezu.

Typical organic acid chromatogram of umezu from different factories was presented in Figure 1 and the main organic acid contents were shown in Table 2. Seven organic acids were detected in the umezu samples, among which six organic acids were identified by HPLC. The major organic acids detected in this assay were citric acid (67.5%–74.3% of total acid) and malic acid (12.9%–18.9% of total acid), and other minor organic acids included oxalic, succinic, lactic, and acetic acid. The organic acid profile of umezu is consistent

TABLE 1: Proximate composition of umezu from different factories.

Umezu samples	pH	Solid content (°Brix)	NaCl (%)	Total acid (%)	Reducing sugar (%)	Total phenolic (mg/L)
Fujian green mume	1.81	34.03 ± 0.04 ^a	22.01 ± 0.54 ^a	5.44 ± 0.24 ^a	8.56 ± 0.03 ^a	300.77 ± 0.37 ^b
Fujian white mume	1.90	33.69 ± 0.02 ^a	21.22 ± 0.15 ^a	5.18 ± 0.11 ^a	7.05 ± 0.05 ^b	250.78 ± 0.19 ^c
Luhe green mume	1.89	31.43 ± 0.11 ^a	20.36 ± 0.48 ^a	4.55 ± 0.15 ^b	5.18 ± 0.02 ^c	260.47 ± 0.85 ^c
Puning green mume	1.85	33.00 ± 0.02 ^a	20.44 ± 0.45 ^a	5.05 ± 0.11 ^a	7.14 ± 0.03 ^b	250.54 ± 0.56 ^c
Raoping green mume	1.86	34.50 ± 0.04 ^a	21.45 ± 0.67 ^a	5.42 ± 0.17 ^a	7.38 ± 0.05 ^b	330.74 ± 0.75 ^a

Means ± SD ($n = 3$) within each column followed by the same letter are not significantly different at the 5% level.

TABLE 2: Predominant organic acids of umezu from different factories (g/L).

Umezu samples	Oxalic acid	Citric acid	Malic acid	Succinic acid	Lactic acid	Acetic acid	Total
Fujian green mume	0.71 ± 0.16 ^a	36.93 ± 0.34 ^b	7.60 ± 0.63 ^b	1.01 ± 0.19 ^a	4.51 ± 0.17 ^a	1.30 ± 0.02 ^a	52.06 ± 1.14 ^a
Fujian white mume	0.36 ± 0.04 ^c	34.06 ± 3.54 ^c	9.54 ± 1.94 ^a	1.13 ± 0.04 ^a	4.20 ± 0.22 ^a	1.15 ± 0.08 ^b	50.44 ± 5.43 ^a
Luhe green mume	0.57 ± 0.06 ^b	30.89 ± 0.73 ^d	6.42 ± 0.12 ^c	1.26 ± 0.24 ^a	3.98 ± 0.20 ^b	0.66 ± 0.21 ^c	43.78 ± 0.73 ^b
Puning green mume	0.46 ± 0.03 ^c	36.27 ± 0.25 ^b	7.59 ± 0.85 ^b	0.90 ± 0.09 ^b	3.83 ± 0.41 ^b	0.95 ± 0.16 ^b	49.99 ± 0.03 ^a
Raoping green mume	0.61 ± 0.13 ^{ab}	38.13 ± 0.76 ^a	6.63 ± 0.16 ^c	1.21 ± 0.21 ^a	3.43 ± 0.81 ^c	1.34 ± 0.06 ^a	51.35 ± 1.75 ^a

Means ± SD ($n = 3$) within each column followed by the same letter are not significantly different at the 5% level.

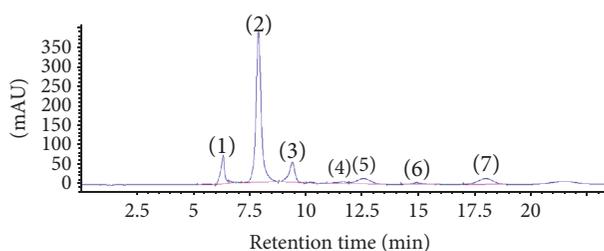


FIGURE 1: Representative chromatogram of organic acid in umezu ($\lambda = 210$ nm). (1) Oxalic acid; (2) citric acid; (3) malic acid; (4) succinic acid; (5) lactic acid; (6) acetic acid; (7) unknown.

with that of *P. mume* fruit reported by others [2, 19]. Citric acid and malic acid are the two most abundant acids, and they account for more than 90% of total acid content in *P. mume* fruit [18] and more than 85% of total acid content in umezu in this study (Table 2). Noteworthily, the differences of these two major organic acids (citric acid and malic acid) among all umezu samples were significant ($p < 0.05$). Except for the different growing environment and species, the fruit maturity might also be a major influence factor [4]. As revealed by Chung et al. [4], malic acid in the fruit is higher than citric acid during the early stage, while citric acid is much more abundant than malic acid during the late maturation stage. Based on Table 2 and Figure 1, it could be concluded that umezu, enriched in organic acids, exhibited potential values in seasoning products development.

3.3. Phenolic Compound Profile. Phenolic compound is another important index for umezu exploitation. Mitani et al. [20] reported that phenolics content in fully matured *P. mume* flesh was as high as 1% on a dry weight basis. Phenolic acids are considered to be anti-inflammatory, anticarcinogenic,

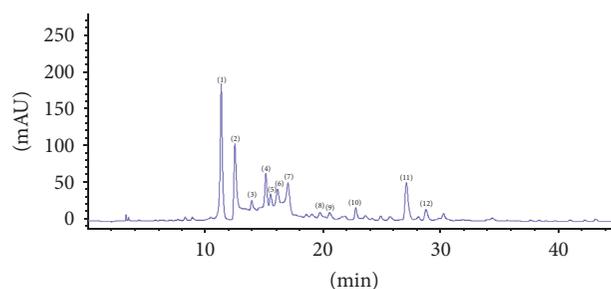


FIGURE 2: Representative chromatogram of phenolic compounds in umezu ($\lambda = 280$ nm). (1) Neochlorogenic acid; (2) unknown; (3) unknown; (4) chlorogenic acid; (5) unknown; (6) unknown; (7) cryptochlorogenic acid; (8) unknown; (9) caffeic acid; (10) unknown; (11) p-coumaric acid; (12) unknown.

and antimicrobial agents, as well as antioxidants [21, 22]. Total and individual phenolic compound contents of umezu samples were shown in Tables 1 and 3, respectively.

Phenolic composition and contents varied in umezu samples. Umezu from Raoping, Guangdong, presented the highest total phenolic content (330.74 mg/L), followed by umezu from Zhaoan, Fujian (300.77 mg/L), while the other three samples had similar total phenolic content. The individual phenolic compounds in umezu samples were assessed by HPLC/DAD and the typical phenolic chromatogram was shown in Figure 2. Five phenolic compounds were identified and quantified, namely, neochlorogenic acid, chlorogenic acid, cryptochlorogenic acid, caffeic acid, and p-coumaric acid. Neochlorogenic acid, chlorogenic acid, and cryptochlorogenic acid accounted for 58.3%–63.6%, 12.4%–18.1%, and 15.4%–20.5% of total quantified phenolic compounds, respectively. In addition, caffeic acid and p-coumaric acid were very low in umezu samples.

TABLE 3: Phenolic composition of umezu of *Prunus mume* from different factories (mg/L).

Umezu samples	Neochlorogenic acid	Chlorogenic acid	Cryptochlorogenic acid	Caffeic acid	p-Coumaric acid
Fujian green mume	161.45 ± 21.99 ^a	39.66 ± 11.99 ^b	39.87 ± 4.55 ^b	3.57 ± 0.39 ^c	9.25 ± 2.49 ^b
Fujian white mume	152.68 ± 33.75 ^b	39.58 ± 14.60 ^b	37.45 ± 7.32 ^b	3.29 ± 0.86 ^c	9.75 ± 3.05 ^b
Luhe green mume	106.64 ± 36.11 ^c	21.91 ± 8.62 ^c	36.18 ± 9.28 ^b	1.98 ± 0.02 ^d	9.58 ± 2.35 ^b
Puning green mume	147.48 ± 14.09 ^b	39.08 ± 9.18 ^b	37.13 ± 4.38 ^b	7.04 ± 0.88 ^a	9.26 ± 2.74 ^b
Raoping green mume	163.38 ± 48.04 ^a	50.79 ± 14.21 ^a	45.77 ± 12.28 ^a	4.77 ± 1.36 ^b	15.75 ± 3.62 ^a

Means ± SD ($n = 3$) within each column followed by the same letter are not significantly different at the 5% level.

TABLE 4: Antioxidant activities of umezu from different factories.

Umezu samples	ORAC ($\mu\text{mol TE/L}$)	ABTS ($\mu\text{g/L}$)	FRAP ($\mu\text{g/L}$)	DPPH (EC_{50}) (mg/mL)
Fujian green mume	12298.34 ± 145.66 ^b	104.67 ± 3.05 ^b	176.43 ± 3.89 ^b	66.00
Fujian white mume	7994.96 ± 266.62 ^d	94.52 ± 10.25 ^c	162.09 ± 9.16 ^d	43.67
Luhe green mume	10361.99 ± 173.19 ^c	24.96 ± 62.72 ^d	172.95 ± 2.44 ^c	56.83
Puning green mume	7741.43 ± 99.94 ^d	94.96 ± 25.21 ^c	162.09 ± 4.24 ^d	65.96
Raoping green mume	17433.72 ± 145.66 ^a	195.83 ± 26.44 ^a	186.43 ± 3.39 ^a	81.70

Means ± SD ($n = 3$) within each column followed by the same letter are not significantly different at the 5% level.

Mitani et al. [20] identified 5-O-caffeoylquinic acid (neochlorogenic acid) and 3-O-caffeoylquinic acid (chlorogenic acid) from the *P. mume* flesh, indicating that the majority of phenolics in *P. mume* fruit were hydroxycinnamic acid derivatives. Xia et al. [23] identified three chlorogenic acid isomers, namely, 3-O-caffeoylquinic acid, 5-O-caffeoylquinic acid, and 4-O-caffeoylquinic acid (cryptochlorogenic acid), from *P. mume* seeds and found 3-O-caffeoylquinic acid was of the highest level in these three isomers. In this study, we also detected the chlorogenic acid isomers in *P. mume* seeds but did not find them (data not shown). According to literatures and our analysis, it is likely that chlorogenic acid isomers in umezu were from *P. mume* flesh, not seeds. During pickling, phenolic compounds in *P. mume* flesh could permeate into umezu. Chlorogenic acid isomers are stable under acidic pH; therefore, they kept a high concentration after pickling was finished. Low level of caffeic acid and p-coumaric acid in umezu might be hydrolyzate of chlorogenic acid isomers. Narita and Inouye [15] reported that chlorogenic acid could hydrolyze into phenolic acids, such as caffeic acid, cis/trans-p-coumaric acid, and ferulic acid.

3.4. Antioxidant Activities. *P. mume* fruits contain multiple antioxidants, including organic acids, isoflavones, and phenolic acids [24]. Antioxidant activities of umezu samples were compared in terms of ORAC, ABTS⁺, FRAP, and DPPH[•] scavenging capacity (Table 4).

Umezu obtained from Fujian white mume and Puning green mume had similar antioxidant properties in terms of ORAC and ABTS⁺ scavenging as well as FRAP. In all antioxidant activity assays, umezu acquired from Raoping green mume and Fujian green mume, however, exhibited significantly higher radical scavenging capacity and reducing power than other samples. Particularly, umezu that came from Raoping green mume presented almost twofold

antioxidant capacity compared with those from Fujian white mume and Puning green mume in ABTS and ORAC values. This could be explained by their higher level of antioxidant ingredients, of which total phenolic acids (330.74 mg/L) and organic acids (51.53 g/L) ranked almost first among all umezu samples (Tables 1 and 2). Reflecting such wealth of organic acids and phenolic acids, the reducing power reflected by FRAP value for umezu samples showed high values, ranging from 162.9 $\mu\text{g/L}$ to 186.43 $\mu\text{g/L}$ Trolox equivalent, depending upon the species and origin areas. In accordance with our results, Debnath et al. [24] revealed that the extract of *Mume Fructus* exhibited high scavenging activities on DPPH, ABTS, hydroxyl, and superoxide radicals with the IC_{50} of 0.40, 0.36, 1.75, and 1.60 mg/ml, respectively. It was reported that the reducing power properties are generally associated with the presence of reductones, which have been shown to exert an antioxidant action by breaking the free radical chains by donating a hydrogen atom [25].

Noteworthy, in all antioxidant activity assays, only ORAC and FRAP of the umezu followed the same trends as their phenolic acids contents; ABTS⁺ and DPPH[•] scavenging capacity went different. Luhe green mume and Raoping green mume presented the lowest and strongest scavenging activities of ABTS⁺, with which ABTS values being 24.96 $\mu\text{g/L}$ and 195.83 $\mu\text{g/L}$, respectively. The DPPH[•] scavenging capacity of samples exhibited the descending order: Raoping green mume > Fujian green mume > Puning green mume > Luhe green mume > Fujian white mume. Umezu samples obtained from Fujian green mume and Puning green mume displayed almost equivalent scavenging activity on DPPH[•] radical ($p > 0.05$). The DPPH[•] scavenging ability of umezu samples obtained from Fujian white mume and Luhe green mume was much lower than other samples, but their EC_{50} values were still higher than 40 mg/ml, which may represent its medicinal value [26].

4. Conclusions

Chemical composition and antioxidant activities of umezu from five *P. mume* pickling factories in China were investigated. These samples contained 33.00–34.50° Brix solid matter in which NaCl was 20.36–22.01%. Total titratable acid content was 4.55–5.44% which makes it with very low pH (1.85–1.90), while reducing sugar content was only 5.18–8.56%. Umezu was high in organic acids (>43 g/L) in which six organic acids were identified and the dominant ones were citric acid and malic acid. It also contained abundant phenolic compounds (>250 mg/L) in which the predominant phenolic acids were neochlorogenic acid, chlorogenic acid, and cryptochlorogenic acid. Umezu obtained from different factories also exhibited strong radical scavenging activities and ferrous reduction ability. Total phenolics were partially responsible for the antioxidant activity assessed by ORAC, ABTS, DPPH, and FRAP assays. Considering the results obtained, it might be anticipated that the umezu could be used as sauce or a natural preservative in food and/or pharmaceutical industry with suitable desalting technology.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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References

- [1] R. Tzonev and M. Yamaguchi, "Investigations on some Far-East Prunus species: Phenology," in *International Symposium on Apricot Culture*, I. Karayiannis, Ed., vol. 1 and 2, pp. 239–242, 1999.
- [2] M. Miyazawa, T. Yamada, and H. Utsunomiya, "Suppressive effect of the Sos-inducing activity of chemical mutagen by citric acid esters from *Prunus mume* Sieb. Et Zucc. using the *Salmonella typhimurium* Ta1535/Psk1002 Umu Test," *Natural Product Research (Formerly Natural Product Letters)*, vol. 17, no. 5, pp. 319–323, 2003.
- [3] I. F. Bolarinwa, C. Orfila, and M. R. A. Morgan, "Amygdalin content of seeds, kernels and food products commercially-available in the UK," *Food Chemistry*, vol. 152, pp. 133–139, 2014.
- [4] H.-S. Chung, D.-S. Kim, H.-S. Kim, Y.-G. Lee, and J.-H. Seong, "Effect of freezing pretreatment on the quality of juice extracted from *Prunus mume* fruit by osmosis with sucrose," *LWT—Food Science and Technology*, vol. 54, no. 1, pp. 30–34, 2013.
- [5] X.-T. Yan, W. Li, Y.-N. Sun et al., "Identification and biological evaluation of flavonoids from the fruits of *Prunus mume*," *Bioorganic & Medicinal Chemistry Letters*, vol. 24, no. 5, pp. 1397–1402, 2014.
- [6] N. Shirasaka, R. Takasaki, and H. Yoshizumi, "Changes in lyoniresinol content and antioxidative activity of pickled Ume during the seasoning process," *Journal of the Japanese Society for Food Science and Technology-Nippon Shokuhin Kagaku Kogaku Kaishi*, vol. 52, no. 10, pp. 495–498, 2005.
- [7] AOAC, *Official Methods of Analysis of AOAC International*, AOAC International, 17th edition, 2002.
- [8] V. Dewanto, W. Xianzhong, K. K. Adom, and R. H. Liu, "Thermal processing enhances the nutritional value of tomatoes by increasing total antioxidant activity," *Journal of Agricultural and Food Chemistry*, vol. 50, no. 10, pp. 3010–3014, 2002.
- [9] H. A. Eyéghé-Bickong, E. O. Alexandersson, L. M. Gouws, P. R. Young, and M. A. Vivier, "Optimisation of an HPLC method for the simultaneous quantification of the major sugars and organic acids in grapevine berries," *Journal of Chromatography B-Analytical Technologies in the Biomedical and Life Sciences*, vol. 885–886, pp. 43–49, 2012.
- [10] H. Ti, Q. Li, R. Zhang et al., "Free and bound phenolic profiles and antioxidant activity of milled fractions of different indica rice varieties cultivated in southern China," *Food Chemistry*, vol. 159, pp. 166–174, 2014.
- [11] N. J. Miller and C. A. Rice-Evans, "Factors influencing the antioxidant activity determined by the ABTS⁺ radical cation assay," *Free Radical Research*, vol. 26, no. 3, pp. 195–199, 1997.
- [12] F. Biglari, A. F. M. Alkarkhi, and A. M. Easa, "Antioxidant activity and phenolic content of various date palm (*Phoenix dactylifera*) fruits from Iran," *Food Chemistry*, vol. 107, no. 4, pp. 1636–1641, 2008.
- [13] R. F. Zhang, F. X. Zhang, M. W. Zhang et al., "Phenolic composition and antioxidant activity in seed coats of 60 chinese black soybean (*Glycine max* L. Merr.) varieties," *Journal of Agricultural and Food Chemistry*, vol. 59, no. 11, pp. 5935–5944, 2011.
- [14] S.-C. Jo, K.-C. Nam, B.-R. Min et al., "Antioxidant activity of *Prunus mume* extract in cooked chicken breast meat," *International Journal of Food Science and Technology*, vol. 41, pp. 15–19, 2006.
- [15] Y. Narita and K. Inouye, "Degradation kinetics of chlorogenic acid at various pH values and effects of ascorbic acid and epigallocatechin gallate on its stability under alkaline conditions," *Journal of Agricultural and Food Chemistry*, vol. 61, no. 4, pp. 966–972, 2013.
- [16] T. Otsuka, T. Tsukamoto, H. Tanaka et al., "Suppressive effects of fruit-juice concentrate of *Prunus mume* sieb. et zucc. (Japanese apricot, Ume) on helicobacter pylori-induced glandular stomach lesions in Mongolian gerbils," *Asian Pacific Journal of Cancer Prevention*, vol. 6, no. 3, pp. 337–341, 2005.
- [17] E. J. Shin, H. J. Hur, M. J. Sung et al., "Ethanol extract of the *Prunus mume* fruits stimulates glucose uptake by regulating PPAR- γ in C₂C₁₂ myotubes and ameliorates glucose intolerance and fat accumulation in mice fed a high-fat diet," *Food Chemistry*, vol. 141, no. 4, pp. 4115–4121, 2013.
- [18] Z.-G. Chen, B.-T. En, and Z.-Q. Zhang, "Simultaneous determination of eight organic acids in *Fructus mume* by RP-HPLC," *Zhongguo Zhong yao za zhi = Zhongguo zhongyao zazhi = China journal of Chinese Materia Medica*, vol. 31, no. 21, pp. 1783–1786, 2006.
- [19] Y. Yu, G. Xiao, Y. Xu, J. Wu, Y. Zhang, and W. Chen, "Changes of quality in the fruits of *Prunus mume* during deacidification by fermentation with *Lactobacillus fermentum*," *Journal of Food Science*, vol. 80, no. 2, pp. M405–M410, 2015.
- [20] T. Mitani, A. Horinishi, K. Kishida et al., "Phenolics profile of mume, Japanese apricot (*Prunus mume* Sieb. et Zucc.) fruit," *Bioscience, Biotechnology, and Biochemistry*, vol. 77, no. 8, pp. 1623–1627, 2013.

- [21] Y. Konishi, S. Kobayashi, and M. Shimizu, "Transepithelial transport of p-coumaric acid and gallic acid in Caco-2 cell monolayers," *Bioscience, Biotechnology, and Biochemistry*, vol. 67, no. 11, pp. 2317–2324, 2003.
- [22] S. Kamiloglu, C. Grootaert, E. Capanoglu et al., "Anti-inflammatory potential of black carrot (*Daucus carota* L.) polyphenols in a co-culture model of intestinal Caco-2 and endothelial EA.hy926 cells," *Molecular Nutrition & Food Research*, vol. 61, no. 2, Article ID 1600455, pp. 1–11, 2017.
- [23] D. Xia, X. Wu, J. Shi, Q. Yang, and Y. Zhang, "Phenolic compounds from the edible seeds extract of Chinese Mei (*Prunus mume* Sieb. et Zucc) and their antimicrobial activity," *LWT- Food Science and Technology*, vol. 44, no. 1, pp. 347–349, 2011.
- [24] T. Debnath, J. P. Bak, N. B. Samad, H. L. Jin, B. R. Lee, and B. O. Lim, "Antioxidant activity of *mume fructus* extract," *Journal of Food Biochemistry*, vol. 36, no. 2, pp. 224–232, 2012.
- [25] N. Sayari, A. Sila, R. Balti et al., "Antioxidant and antibacterial properties of *Citrus paradisi* barks extracts during turkey sausage formulation and storage," *Biocatalysis and Agricultural Biotechnology*, vol. 4, no. 4, pp. 616–623, 2015.
- [26] C. J. Seneviratne, R. W. K. Wong, U. Hägg et al., "*Prunus mume* extract exhibits antimicrobial activity against pathogenic oral bacteria," *International Journal of Paediatric Dentistry*, vol. 21, no. 4, pp. 299–305, 2011.



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