Evaluation of onion juices quality following heat-treatment and their application as a sugar substitute in Kimchi

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Abstract This study was conducted to evaluate the quality of onion juices that had been heat-treated for different times as well as their use as a table sugar substitute in Kimchi. The onions were steamed at 100 °C for 30 min and boiled at 90 °C for 30, 60, and 120 min. The highest cycloalliin (0.76 mM), free-sugar (sucrose 1.66 g/L, glucose 8.62 g/L, and fructose 7.64 g/L), and malic acid (0.82 g/L) contents were observed in onion boiled at 90 °C for 120 min. The possibility of using heat-treated onion juices as an alternative to table sugar in Kimchi was evaluated by comparing the lactic acid bacteria count, pH, acidity, organic acid, and free-sugar in these juices with those in Kimchi prepared using table sugar (control). The total viable bacteria and lactic acid bacteria showed similar growth patterns as in the control. The average pH reduction and increase in titratable acidity (%) in all treated Kimchi samples during fermentation for 4 weeks were 1.18 ± 0.05 and 0.81 ± 0.06 , respectively. Kimchi with onion juice heat-treated for 120 min (K120) had the most similar lactic acid and acetic acid contents to that in the control after fermentation for 4 weeks. The highest mannitol level after fermentation for 4 weeks was detected in K120, which showed better sensory qualities compared to the control.

Keywords Heat-treatment · Onion · Kimchi · Free-sugar · Organic acid · Lactic acid bacteria

Introduction

Onion (*Allium cepa* L.) is a plant belonging to the *Liliaceae* family that grows naturally and is one of the most widely used edible plants worldwide (Abdelrahman et al. 2016; Griffiths et al. 2002; Shokoohinia et al. 2015; Zhang et al. 2016). In several studies, onion and onion extracts were shown to decrease the level of blood lipid, increase fibrinolysis, decrease platelet aggregation, and lower blood pressure (Saleheen et al. 2004). These biological activities are exerted by phenolic and sulfur compounds (Hovius and Goldman 2005; Kim et al. 2006, 2012; Lee et al. 2005).

Commonly, the vegetables are subjected to heat treatments in commercial processing plants or domestic household kitchens. The degree of changes in the phytochemicals present in edible vegetables during heat-treatments depends on the sensitivity of the phytochemicals to modification or degradation and the duration of exposure to a given processing technique (Breene 1994). Several studies have been conducted to evaluate the changes in phytochemicals in vegetables during cooking (Robards et al. 1999; Kim et al. 2013). However, few studies have evaluated the changes in taste-related compounds, such as free-sugar, organic acid, and sulfur compounds in heat-treated onions or the application of heat-treated onions in fermented foods (Benkeblia 2000; Kim et al. 2012).

Kimchi is a typical Korean fermented food made by fermentation of Napa cabbage with red pepper, onion, garlic, salts, and table sugar (Jung et al. 2012). It is known to have various functions such as anti-cancer, anti-obesity, cholesterol reduction, and antioxidant effects (Park et al.

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2014). The high intake of table sugar is added to improve the taste and facilitate the fermentation of Kimchi (Park et al. 1995), but it has negative health effects. The intake of high table sugar can lead to diabetes, cancer and cardiovascular disease (Baynes and Thorpe 1999; Giugliano et al. 1996; Robertson et al. 2004). Therefore, the development of table sugar substitute is necessary for reducing the intake of sugar. Previous studies have shown that heat-treatment increase the content of free-sugar and sulfur in onions (Kim et al. 2016; Woo et al. 2007).

Therefore, the objective of this study was to improve the free-sugar, organic acid and cycloalliin contents of onions juices by heat-treatment, as well as to evaluate whether heat-treated onion juice can be used as a substitute for table sugar in Kimchi.

Materials and methods

Materials

Onions, garlic, ginger and Napa cabbage were purchased from the market in Gwangju, Korea. Sun-dried salt and fish-sauce purchased from Cheiljedang Co. (Seoul, Korea). Red pepper powder (Imsil Red Pepper Co., Imsil, Korea), fish sauce (Chungjungone Co., Seoul, Korea), glutinous rice (Dami-food Co., Gwangju, Korea) and table sugar (Samyang Co., Ulsan, Korea) were purchased from the markets. Cycloalliin, which was used as the standard for analysis was purchased from Wako Co. (Wako Pure Chemical Industries, Ltd., Osaka, Japan), Free-sugar standards (sucrose, glucose, fructose, and mannitol) and organic acid standards (citric acid, malic acid, lactic acid, fumaric acid, and acetic acid) purchased from Duksan (Seoul, Korea). All the chemicals and solvents used were of analytical grade.

Preparation of heat-treated onion juices

Heat-treated onion was prepared as described by Kim et al. (2016) and Woo et al. (2007), with some modifications. Briefly, non-damaged onions, weighing 160–220 g, were peeled and washed. The fresh onions were steamed at 100 °C for 30 min and boiled at 90 °C for 30, 60, 120, and 240 min. The heat-treated onions were homogenized using a blender (Hanil, Seoul, Korea) for 5 min, and the homogenate was filtered through a Whatman filter paper (Whatman No. 2, Maidstone, UK). The prepared onion juice was stored at − 20 °C until analysis.

Preparation of Kimchi

Kimchi was prepared using salted Napa cabbage and seasoning mixtures. The seasoning mixtures was composed of red pepper powder, garlic, ginger, fish sauce, glutinous rice, water, table sugar, salts and different heat-treated onion juices. The seasoning mixtures were mixed with salted Napa cabbage at a ratio 3:7 and stored at 4 °C for 4 weeks. The following Kimchi groups were prepared: (i) Control: Kimchi containing red pepper powder, garlic, ginger, fish sauce, glutinous rice, water, table sugar, and salts; (ii) K30: table sugar-free Kimchi containing onion juice heat-treated for 30 min; (iii) K60: table sugar-free Kimchi containing onion juice heat-treated for 60 min; and (iv) K120: table sugar-free Kimchi containing onion juice heat-treated for 120 min.

Analysis of cycloalliin

The cycloalliin content in Kimchi and heat-treated onion juice was analyzed using an Agilent 1260 HPLC system (Agilent Technologies, Santa Clara, CA, USA) coupled with a quadrupole mass spectrometer Agilent 1000 (Agilent Technologies). The HPLC condition for quantitation of cycloalliin was performed according to Kim et al. (2016) with some modifications. The samples were injected into an Aegispak C-18 column (2 × 150 mm, 3 µm pore size). A gradient elution system was employed using mobile phase A and B. Mobile phase A consisted of H₂O containing 0.2% (v/v) phosphoric acid, and mobile phase B consisted of acetonitrile containing 0.2% phosphoric acid. The gradient system was as follows: 0–5 min, B 84%; 5.1–15 min, B 80%; 15.1–20 min, B 84%. The injection volume was 20 µL and flow rate was 0.2 mL/min. The column temperature was set to 25 °C. The mass spectrometry was operated in a positive ionization mode for sensitive and selective detection using multiple reaction monitoring mode. Positive ionization was performed at an ion-spray voltage of 5000 V, curtain gas of nitrogen at 20 psi, ion source gas 1 and 2 at 50 psi, and temperature of 550 °C. The mass parameters of precursor ion, product ion, declustering potential, entrance potential, collision energy, and collision cell exit potential were optimized by infusing standard stock solutions (cycloalliin, 1 µg/mL). The declustering potential was 53.0 V, entrance potential was 10.0 V, collision energy was 23.0 V, collision exit potential was 10.0 V for cycloalliin.

Analysis of free-sugar

The free-sugar content of Kimchi and heat-treated onion juice were determined according to Benkeblia et al. (2002) with some modifications. The samples (5 g) were mixed

with 50 mL water and extracted in a water bath at 85 °C for 25 min and then cooled at 25 °C. The extract solutions was filtered through 0.2-µm filter and stored at − 40 °C before high-performance liquid chromatography (HPLC) analysis.

The contents of free-sugar in Kimchi and heat-treated onion juice were determined using an Agilent 1260 HPLC system (Agilent Technologies). A refractive index (RI) detector and carbohydrate column (Sugar-pak I Column, Waters, Milford, MA, USA) were used. The mobile phase (acetonitrile: water = 80:20 v/v) was used at a flow rate 1 mL/min with isocratic mode. The injection volume was 20 µL, and the column oven was maintained at 30°C. Sucrose, glucose, fructose, and mannitol were used as standard compound. For each sugar, determination was done three times.

Analysis of organic acid

Organic acid in Kimchi and heat-treated onion juice were analyzed as described by Gao et al. (2012) with some modifications. The samples (1 g) were mixed with distilled water and extracted using a probe-type ultrasonicator (Sonics & Materials, Newtown, CT, USA) for 4 min. The extracts solutions were filtered through 0.2 µm filters and stored at − 40 °C before HPLC analysis.

Organic acids were determined by chromatography on an Agilent 1260 HPLC system equipped with an Aminex HPX-87H column (300 × 7.8 mm, 9 µm, Bio-Rad, Hercules, CA, USA) kept at 50 °C. The injection volume was 20 µL. Isocratic elution was performed with 0.01 N H₂SO₄ in deionized water as the mobile phase for 30 min at a flow rate of 0.8 mL/min. A variable wavelength detector (DAD) at a wavelength of 210 nm was used for determine the organic acid contents. The organic acids in the samples were identified by comparing their retention times with those of standard organic acids and were quantified using a calibration curve derived from the peak areas of the standards. Citric acid, malic acid, lactic acid, fumaric acid, and acetic acid used as standards. For each sample, was done three times.

Analysis of pH and acidity

Kimchi sample (10 g) was mixed with distilled water (100 mL) and homogenized using a blender (Hanil) for 5 min; the homogenate was then filtered (Whatman filter paper No. 2, Maidstone, UK). The pH of Kimchi juice was measured using a pH meter (Thermo Electron Corporation, Waltham, MA, USA). The titratable acidity (%) of each sample was measured following the standard AOAC protocol (AOAC 2000). The Kimchi juice was titrated to pH 8.3 by adding 0.1 N NaOH and the titratable acidity was

calculated as a percentage of lactic acid (MW = 90.08) by using the following formula:

$$\text{Titratable acidity (\%)} = \frac{A \times N \times 90.08 \times 100}{V \times 1000},$$

where *A* is the volume of 0.1 NaOH (mL) used, *N* is the normality of 0.1 N NaOH (mL), 90.08 is molecular weight, and *V* is the mass (g) of the sample used.

Microbial analysis

Kimchi sample (10 g) was mixed with 0.85% NaCl (100 mL) in a sterile filter bag and homogenized for 1 min using a stomacher (Bagmixer R400, Interscience, Saint Nom, France). The total viable bacteria count was determined using plate count agar (Difco, Detroit, MI, USA) after incubation at 36 °C for 48 h. Lactic acid bacteria levels were determined using de Man, Rogosa and Sharpe (Difco) agar after incubation at 36 °C for 72 h. Total colony counts were determined in triplicate and the results were expressed as log CFU/g.

Sensory analysis

Sensory evaluation of Kimchi was conducted according to Jung et al. (2012) with some modifications. Kimchi sample was stored at 4 °C in a refrigerator and sensory evaluation was performed after 4 weeks. Kimchi samples were randomly presented in a clear plastic dish with a 3-digit code number to 15 trained panelists. Water and crackers were provided for the panel to rinse their palates between samples. Sensory evaluation (sweetness, bitterness, saltiness, spicy, off-flavors, flavor preference, and overall acceptability) was scored on a 5-point rating scale, where 5 = very good; 4 = good; 3 = acceptable; 2 = poor; and 1 = very poor.

Statistical analysis

All measurements were performed in triplicate, and all data were expressed as the means ± standard deviation. Statistical analysis was performed using SPSS version 20.0 software (SPSS, Inc., Chicago, IL, USA). Significant differences among treatments were determined using one-way analysis of variance (ANOVA) and Duncan's multiple range test. The level of significance was set to *p* < 0.05.

Results and discussion

Characteristics of heat-treated onion juices

The quality of heat-treated onion juice was determined by evaluating the content of cycloalliin, free-sugar and organic acid. Figure 1 was shown that the longer extraction time lead to an increase in cycloalliin content of the onion juices. Cycloalliin is also one of the major sulfur compounds present in onion (*Allium cepa* L.) and is known to have several biological effect such as anti-arteriosclerosis and anti-cancer. However, these compounds are unstable and are transformed to different products. For this reason, attention has recently been focused on polar compounds that are more stable during cooking and under storage (Lanzotti 2006). The results obtained in the present study were consistent with those of previous studies, where it was shown that the cycloalliin content of onion extracts was increased by heat-treatments and the amount of cycloalliin was the highest in steaming process (Kim et al. 2016; Ueda et al. 1994). The highest cycloalliin (0.76 ± 0.05 mM) was present in onion heat-treated at 90 °C for 120 min. This conditions were increased the cycloalliin by about 58% compared to the non-heated onion juice.

Onion juices included free-sugars, such as sucrose, fructose and glucose (Davis et al. 2007). The content of free-sugar in heat-treated onion juices was increased upon heat-treatment (Fig. 2). The onion heat-treated at 90 °C for 120 min the highest content of sucrose (1.66 ± 0.00 g/L), glucose (8.62 ± 0.01 g/L) and fructose (7.64 ± 0.01 g/L). The heat treatments (boiling and steaming) were reported to result in the released hydrophilic free-sugar from onion (Kim et al. 2012; Lee and Jung 2012). Davis et al. (2007) was showed the effect of hot water extraction in improving the concentration of free-sugars. As the extraction time was increased, there was an increase in the content of organic

Fig. 1 Effect of heat-treatment on cycloalliin (mM) content in onion juices. Different letters indicate a significant difference at $p < 0.05$ ($n = 3$) according to Duncan's multiple range test

Fig. 2 Effect of heat-treatment on the content of free sugar (g/L) in onion juices. **a** Sucrose, **b** glucose, and **c** fructose. Different letters indicate a significant difference at $p < 0.05$ ($n = 3$) according to Duncan's multiple range test

acids, but not in that of cycloalliin, and the content of free-sugars was decreased. The effect of heat treatment on the content of organic acid of onion juice is shown Fig. 3. The malic acid was increased with upon heat treatment (Fig. 3a). The onion heat-treated at 90 °C for 120 min had the highest citric acid (0.23 ± 0.00 g/L) (Fig. 3b) and fumaric acid (0.03 ± 0.00 g/L) contents (Fig. 3d). The content of lactic acid and acetic acid were not influenced by heat treatment (Fig. 3c, e). These results were observed because malic acid, citric acid, fumaric acid is released from the vacuoles of onion cells and lactic acid and acetic acid (Lee and Jung 2012; Lee et al. 2011; Choi et al. 2014). Additionally, Kim et al. (2014) reported that higher organic acid contents from onion was obtained by steaming and boiling rather than by heat treatment. These results indicate

Fig. 3 Effect of heat treatment on the content of organic acid (g/L) in onion juices. **a** Malic acid, **b** citric acid, **c** lactic acid, **d** fumaric acid, and **e** acetic acid. Different letters indicate a significant difference at $p < 0.05$ ($n = 3$) according to Duncan's multiple range test

that heat treatment at 90 °C for 120 min is effective for increasing sulfur compound, free-sugar, and organic acid levels in onion juice.

Physicochemical properties of Kimchi with heat-treated onion juices

The potential for using heat-treated onion juices as an alternative to sugar in Kimchi was evaluated by counting the lactic acid bacteria and by measuring organic acid (lactic acid and acetic acid) and free-sugar (mannitol and sucrose) contents with those in the control.

Table sugar is used as a feed for growth of lactic acid bacteria in Kimchi (Cho et al. 2006; Cheigh et al. 1994). Total viable bacterial and lactic acid bacterial counts were similar to those in the control (Fig. 4a, b). Additionally, the growth of lactic acid bacteria was associated with a decreased pH and increased titratable acidity (%) in Kimchi. The average pH reduction and titratable acidity

increase in all treated Kimchi samples during fermentation for 4 weeks (Fig. 4c, d).

According to a previously study, lactic acid bacteria produce specific organic acids and free sugars during fermentation (Cho et al. 2006). Therefore, changes in the contents of specific organic acids (lactic acid and acetic acid) and free-sugars (mannitol and sucrose) were evaluated. Lactic acid and acetic acid levels increased with fermentation time in all treated Kimchi samples. The content of lactic acid and acetic acid were rapidly increased during the early stage of fermentation; the highest lactic acid and acetic acid levels were observed in the control. Kimchi with onion juice heat-treated for 120 min (K120) showed the most similar lactic acid and acetic acid contents as the control after fermentation for 4 weeks (Fig. 4e, f).

Lactic acid bacteria (*Lactobacillus* sp. and *Leuconostoc* sp.) produce mannitol under fermentation conditions (Saha and Nakamura 2003; Soetaert et al. 1995). After fermentation for 2 weeks, the sucrose in Kimchi, containing heat-treated onion juices was consumed by lactic acid bacteria

Fig. 4 Change in **a** total viable bacteria, **b** lactic acid bacteria, **c** pH, **d** titratable acidity, **e** lactic acid, and **f** acetic acid in Kimchi with heat-treated onion juices during fermentation. Diamond: Con is normal Kimchi containing red pepper powder, garlic, ginger, fish sauce, glutinous rice, water, table sugar, and salted cabbages; open circle: K30 is table sugar-free Kimchi with onion juice heat-treated for 30 min; filled circle: K60 is table sugar-free Kimchi with onion juice heat-treated for 60 min; and filled triangle: K120 is table sugar-free Kimchi with onion juice heat-treated for 120 min. All data are the average value $\pm$ standard deviation determined from triplicate measurements

(Fig. 5a), whereas mannitol content was increased after fermentation for 2 weeks (Fig. 5b). After fermentation for 4 weeks, the highest mannitol content was observed in K120. These results indicate that K120 can replace table-sugar for the growth of lactic acid bacteria.

Sensory evaluation of Kimchi with heat-treated onion juices

The purpose of this study was to determine whether heat-treated onion juice could be used as a substitute for sucrose in Kimchi. K120 was selected as an optimized Kimchi with heat-treated onion juice in this study. Sensory evaluation was conducted by comparing K120 to the control. The

prepared Kimchi samples were stored at 4 °C and sensory evaluation was performed after 4 weeks. The sweetness, flavor preference, bitterness, and overall acceptability of K120 were higher than those of the control. The saltiness, spicy, and off-flavors of K120 were lower than those of the control (Fig. 6a, b). The sweetness of control and K120 scored 2.2 and 2.4, respectively. Also, overall taste and flavor of K120 was higher than control. These results indicated that the contents of free-sugar and organic acid in onion were increased by heat treatment because of the sugar-alcohol produced during fermentation of Kimchi.

In a previously study, the effect of fresh onion on Kimchi taste was evaluated and found to the flavor of Kimchi was found to improve when an appropriate amount

Fig. 5 Change in **a** sucrose, and **b** mannitol on kimchi with heat-treated onion juices during fermentation. Filled square: Con is normal Kimchi containing red pepper powder, garlic, ginger, fish sauce, glutinous rice, water, table sugar, and salted cabbages; gray filled square: K30 is table sugar-free Kimchi with onion juice heat-treated for 30 min; dashed line square: K60 is table sugar-free Kimchi with onion juice heat-treated for 60 min; and dotted line square: K120 is table sugar-free Kimchi with onion juice heat-treated for 120 min. Different letters indicate a significant difference at $p < 0.05$ ($n = 3$) according to Duncan's multiple range test

of fresh onion was added (Choi and Cho 2009). Additionally, Rajeswari et al. (2014) reported that addition of onion powder during pasta processing improved quality and sensory qualities.

Kimchi contain lactic acid bacteria and heat-treated onions contains metabolites such as organic acid, free-sugar and sugar alcohol. Base on the results of sensory evaluation, the sensory properties were not decreased by using heat-treated onions as a substitute for in Kimchi.

Conclusion

Vegetables are subjected to heat-treatments in commercial processing plants or household kitchens. The degree of phytochemical changes during heat treatments depends on the sensitivity of a phytochemical to modification or degradation and the length of exposure time to a given processing techniques. Thus, many studies have been conducted to evaluate the changes in phytochemicals in

Fig. 6 Radar plots of the sensory evaluation of kimchi at 5 °C after 4 weeks of fermentation. **a** Con and **b** K120. Con is normal kimchi containing red pepper powder, garlic, ginger, fish sauce, glutinous rice, water, table sugar, and salted cabbage; K120 is table sugar-free kimchi with onion juice heat-treated for 120 min

vegetables during cooking. However, few studies have evaluated the changes in taste-related compounds of heat-treated onions, such as free-sugar, organic acid, and sulfur compounds or the application of heat-treated onions in fermented foods. Therefore, the objective of this study was to improve the free-sugar, organic acid and cycloalliin contents of onions juices by heat treatment, as well as to evaluate whether the heat-treated onion juice can be used as a substitute for sugar in Kimchi. As shown in our results, heat treatment at 90 °C for 120 min was effective in increasing the contents of sulfur compound, free- sugar, and organic acid levels in onion juice and K120 can replace for sugar in the growth of lactic acid bacteria. Also, the sensory properties were not decreased by using heat-treated onions as a substitute in Kimchi. Based on these results, it is suggested that heat-treated onion juice can replace table sugar in fermented food.

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Compliance with ethical standards

Conflict of interest The authors declare no conflicts of interest.

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