

# Chemical, nutritional, and bioactive compositions of fresh, washed, and blanched shiitake

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**Citation:** Mau J.L., Tseng J., Wu C.R., Chen C.H., Lin S.D. (2021): Chemical, nutritional, and bioactive compositions of fresh, washed, and blanched shiitake. Czech J. Food Sci., 39: 000–000.

**Abstract:** Washing and blanching are two important steps for the processing of fresh good grade small shiitake into casual snacks and prepared foods. This study explored their effects on the dimensions, weights, proximate compositions, taste and bioactive compounds of the shiitake caps and stipes. Results showed that the dimensions and weights of the caps and stipes increased after washing but decreased after blanching. On a dry basis, washing did not affect the nutrients and bioactive compounds contents of shiitake except sugars and polyols compared to fresh samples. However, blanching greatly reduced all the quality contents of the shiitake. The major minerals in the caps and stipes were potassium, magnesium and calcium. The main polyol, sugar, free amino acid, and 5'-nucleotide of shiitake were mannitol, trehalose, glutamine and 5'-adenosine monophosphate, respectively. Equivalent umami concentration (EUC) in the caps was three-fold higher than that in the stipes. Ergosterol, ergothioneine,  $\gamma$ -aminobutyric acid (GABA), and total phenols contents in the caps were significantly higher than those of the stipes, but the polysaccharides showed the reversed. Overall, both caps and stipes contain the necessary nutrients and bioactive compounds, and the blanching significantly affected the above-mentioned components of all tested samples.

**Keywords:** *Lentinula edodes*; washing; blanching; nutrient; bioactive compound

*Lentinula edodes* (Berk.) Pegler, called shiitake in Japan and Xianggu in China, is one of the most important traditional edible and medicinal fungi in Asian countries. It consists of a cap and stipe that has a unique flavour, texture and medicinal properties and is preferred by consumers (Zhang et al. 2007; Sun et al. 2020). Shiitake contains proximate compositions, minerals, soluble sugars and polyols, free amino acids (AA), 5'-nucleotides and volatile compounds (Yang et al. 2001; Ulziijargal and Mau 2011; Cohen et al. 2014; Chen et al. 2015; Li et al. 2017, 2018; Sun et al. 2020). It also contains a number

of functional bioactive compounds, such as ergothioneine, ergosterol,  $\gamma$ -aminobutyric acid (GABA), polysaccharides and total phenols (Yang et al. 2002; Zhang et al. 2007; Chen et al. 2012; Cohen et al. 2014).

In Taiwan, fresh shiitake are graded as special, premium, and good and are divided into three different sizes according to their cap diameters: small (3–5 cm), medium (5–7 cm) and large (> 7 cm). Among them, good grade shiitake with small size has low commodity prices in the market, so they were channelled into processed mushrooms, such as vacuum-fried crisps.

Supported by the Ministry of Science and Technology and Mei Tong Co. Ltd. of Taiwan (Projects No. MOST 107-2622-E-241-001-CC3 and No. HK106-023).

After the shiitake are delivered to the food factory, the stipes are removed at the first processing step. Thereafter, washing and blanching are carried out, which are two important steps for processing shiitake into vacuum-fried crisps and prepared foods. The main purpose of washing is to remove the foreign bodies on the surface of shiitake, while the main purpose of blanching is to destroy the enzyme activity and avoid shiitake deterioration. Besides, hot water blanching is one of the most widely employed traditional pretreatment methods and could be used to reduce or eliminate the bitterness of vegetables.

There are few relevant literature sources (Li et al. 2017) on the effects of washing and blanching steps on the quality characteristics of shiitake. Accordingly, the primary goal of this research is to examine the changes of dimensions, weights, chemical compositions, nutritional and medicinal values of shiitake caps and stipes during washing and blanching.

## MATERIAL AND METHODS

**Material and chemicals.** The small size of fresh good grade shiitake was purchased from a mushroom farm in Nantou, Taiwan. Gallic acid, Folin-Ciocalteu's phenol reagent (2 N), sugars and polyols, AA and 5'-nucleotides, GABA, ergosterol, ergothioneine, sodium dodecyl sulfate, trimethylamine, betaine, 2-mercapto-1-methylimidazole, sodium acetate, tetrahydrofuran, 1,4-dithiothreitol were purchased from Sigma-Aldrich (St. Louis, Missouri, USA). Minerals were purchased from AccuStandard, Inc. (New Haven, Connecticut, USA). Methanol (99.9%) and acetonitrile (99.9%) were purchased from Avantor Performance Materials (Randor, Pennsylvania, USA). Hexane (99.9%) was purchased from Tedia (Fairfield, Ohio, USA). All other chemical reagents used were of analytical grade, and the purity is greater than 98%, except hydrogen peroxide (30%) and nitric acid (70%).

**Preparation of tested samples.** For the measurement of diameters, lengths and weights of fresh shiitake (approximately 3 kg), scissors were used to separate the caps and stipes, followed by washing (30 s, running water) and blanching [5 min, 1 : 5 ratio of sample to boiling water ( $98 \pm 2^\circ\text{C}$ )]. Fresh, washed and blanched caps were designated as FC, WC, and BC, respectively, whereas fresh, washed and blanched stipes were designated as FS, WS, and BS, respectively. Thereafter, fresh shiitake (45 kg) were separated with scissors to separate the caps and stipes and were divided into three batches. The first, second, and third batch samples were used as fresh,

washed, and blanched samples, respectively. All samples were lyophilised and were ground and screened in a mill with 0.2 mm sieve (RT-30HS; Rong Tsong Precision Technology, Taichung, Taiwan). All powder samples obtained were sealed in PET/Al/PE bags (Sun A Enterprise Co., Ltd., Taichung, Taiwan) and stored in a freezer ( $-25^\circ\text{C}$ ) before use (NL; Cygnus, Inc., Taipei, Taiwan).

**Determination of dimensions, weights, proximate compositions and minerals.** The dimensions and weights of the samples were measured with a calliper (500-196-20; Mitutoyo, Kanagawa, Japan) and a scale (AB204-S; Mettler Toledo, Zurich, Switzerland), respectively. Ash, fat, moisture, and protein of the samples were determined by AACC International (2000) Approved Methods (08-01.01, 30-25.01, 44-40.01, and 46-11.02, respectively). The nitrogen conversion factor of the sample used for crude protein content calculation is 4.38 (Ulziiyargal and Mau 2011). Carbohydrate content (%) was calculated by subtracting the contents of protein, fat and ash from 100% of matter on a dry basis. Minerals were determined following the method of test for minerals in infant formula – test of copper, iron, magnesium, manganese, potassium, sodium, and zinc [National Standards of the Republic of China, general No. 12869, classified No. N6231, Bureau of Standards, Metrology and Inspection, Ministry of Economic Affairs (MOEA), Taiwan] described in CNS (2018) and were analysed by high-resolution inductively coupled plasma mass spectrometry (ICP-MS) (Thermo Scientific<sup>TM</sup> Element 2<sup>TM</sup>; Thermo Fisher Scientific Inc., Waltham, Germany).

**Determination of taste-related compounds.** Sugars, polyols and 5'-nucleotides were determined following the method of Tsai et al. (2007). Free AA was determined following the methods used in Hou et al. (2018). Equivalent umami concentration (EUC) [ $\text{g monosodium glutamate (MSG)} 100 \text{ g}^{-1}$ ] was calculated on the basis of the addition equation established in Yamaguchi et al. (1971).

**Determination of bioactive compounds.** Ergothioneine and GABA were determined following the methods described previously (Chen et al. 2012). Ergosterol was analysed following the method used in Qian and Sheng (1998). Crude polysaccharides and total phenols were measured according to the methods of Mau et al. (2019).

**Statistical analysis.** Each measurement was conducted in triplicate, except for the dimensions and weights ( $n = 326$ ). All data were analysed using analysis of variance followed by Duncan's multiple range tests to determine significant differences among the means at the level of  $\alpha = 0.05$  (version 9.3; SAS Institute, Cary, North Carolina, USA).

## RESULTS AND DISCUSSION

**Dimensions and weights of shiitake.** The 96.62% of the FC (average 38.37 mm) falls within 30–50 mm diameter criteria (Figure 1). The thickness of FC (average 12.08 mm) accounts for 92.03% in the range of 8–16 mm. The weight of FC (average 7.51 g) accounts for 71.47% between 6–10 g. After washing, the diameter, thickness, and weight of WC grew to 39.56 mm, 12.23 mm, and 11.35 g on average, respectively. The average diameter and thickness of BC are 33.31 mm and 8.64 mm, respectively, which were significantly lower than FC, but its average weight (7.89 g) increased, which may be due to water retention. The average diameter, length and weight of the FS are 11.27 mm, 21.19 mm, and 1.64 g, respectively (Figure 2). After washing, the average diameter, length and weight of the WS increased to 12.20 mm, 21.76 mm, and 2.03 g, respectively. Blanching reduced the stipes diameter to an average of 10.92 mm, while the length increased to an average of 22.89 mm. The weight of the BS averaged 1.71 g.

**Proximate compositions and minerals.** Both the caps and stipes gained moisture during the washing process (Table 1). Based on drying, the results show that the protein, fat, ash, and carbohydrate contents of the sample are not affected by washing. After blanching, the protein, fat, and ash contents of the sample significantly decreased, while carbohydrate showed the reversed. This is because during the blanching process, the protein, fat and ash of the sample are leaking into the blanching water higher than the carbohydrate. The protein, fat, and ash contents in the FC were significantly higher than those in the FS, while carbohydrate showed the reversed. The results of Ulzijargal and Mau (2011) and Li et al. (2018) demonstrated that the carbohydrate content of stipes is higher than that of caps, while the protein, fat, and ash contents are the opposite, similar to the results of this study.

Of the major minerals, Mg, P, K, and Na contents were all significantly higher in the caps than the stipes (Table 2); however, Ca was significantly higher in the stipes than the caps. The major mineral for both caps and stipes is K. The results were consistent with the findings of Cohen et al. (2014) and Li et al. (2018). Among the trace minerals, Al, Cr, Mn, and Se were all higher in the stipes than in the caps. The Cd and Pb content of samples in this study did not exceed the legal limits ( $\text{Cd} < 2 \text{ mg kg}^{-1}$  dry matter and  $\text{Pb} < 3 \text{ mg kg}^{-1}$  dry matter) set by the Taiwan Food and Drug Administration. Washing does not affect the mineral content of shiitake but

blanching reduces its mineral content. This might be due to the fact, that blanching destroyed the cell structure, causing the release of minerals into the blanching water.

**Sugars and polyols.** Mannitol, arabitol, trehalose and glucose were the main polyols and sugars in shiitake. The content of mannitol was highest in the caps, and the arabitol value was the highest in the stipes (Chen et al. 2015). In this study, arabinose, fructose, and glucose of samples were not detected. Mannitol and trehalose concentrations were significantly higher in the caps than those in the stipes, but arabitol showed the reversed (Table 3). Total sugars and polyols content in the stipes were significantly higher than those in the caps. This result is similar to that of Chen et al. (2015). Washing and blanching reduced the sugars and polyols content of the shiitake. This result is similar to that of Li et al. (2017), who pointed out that hot water blanching will reduce the sugars and polyols content of shiitake. Glucose was not detected in the sample, which might be due to the difference in size and cultivation media of shiitake.

**Free amino acids (AA).** Except for leucine in the stipes, washing did not affect the AA content in samples, but blanching can significantly reduce the AA content (Table 4). Glutamine, methionine, glutamic acid, and arginine are the major AA in the caps. All essential AA were found in the FC, WC, and BC and constituted about 37%, 37%, and 41% of total AA contents, respectively. Branched-chain AA (BCAA, leucine, isoleucine and valine) constituted about 23%, 22%, and 21% of the essential AA in the FC, WC, and BC, respectively. Major AA found in the stipes were glutamine, alanine, tryptophan, arginine, glutamic acid, and valine. Except for methionine, other essential AA were detected in the stipes. The BCAA constituted about 35%, 35% and 25% of the essential AA in the FS, WS, and BS, respectively.

The AA are grouped based on their taste characteristics, MSG-like (aspartic acid and glutamic acid), sweet (alanine, glutamine, glycine, proline, serine, and threonine), bitter (arginine, histidine, isoleucine, leucine, methionine, phenylalanine, tryptophan, and valine) and tasteless (lysine and tyrosine) (Yang et al. 2001). MSG-like AA were 1.88- and 1.95-fold higher in the caps than in the stipes for fresh and washed samples, respectively, but only 1.21-fold higher in the blanched samples. The sweet AA content of the stipes was higher than that of the caps, but the bitter and tasteless AA showed the reversed. Bitter AA have a higher proportion in samples, but it can be masked by the sweet AA, sugars, and polyols. In this study, the MSG-like AA content of FC and FS were lower than those of Li et al. (2018) (36.18–77.88  $\text{mg g}^{-1}$ ) and

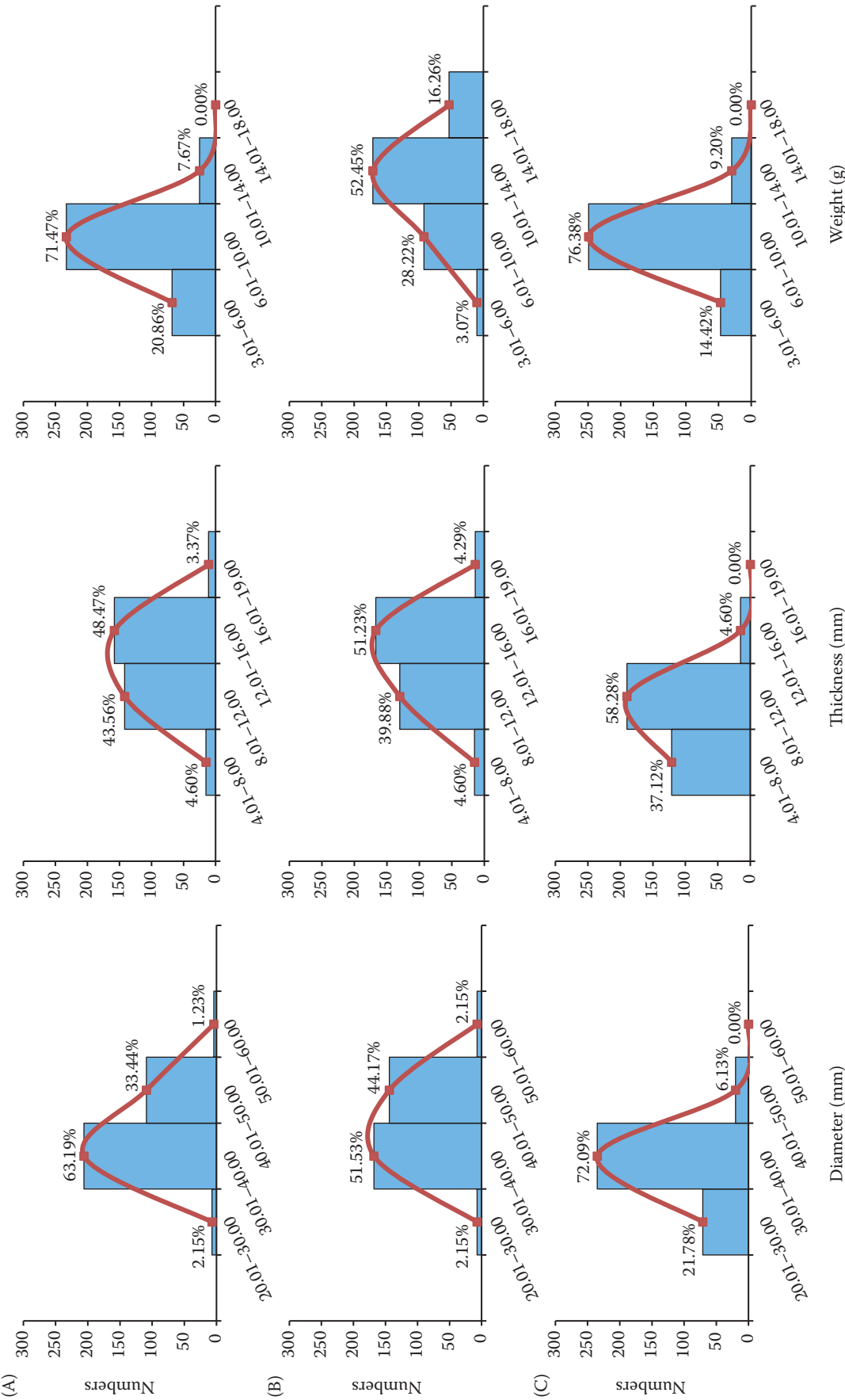


Figure 1. Effects of washing and blanching on the dimensions and weights of shiitake caps: (A) fresh caps, (B) washed caps and (C) blanched caps ( $n = 326$ )

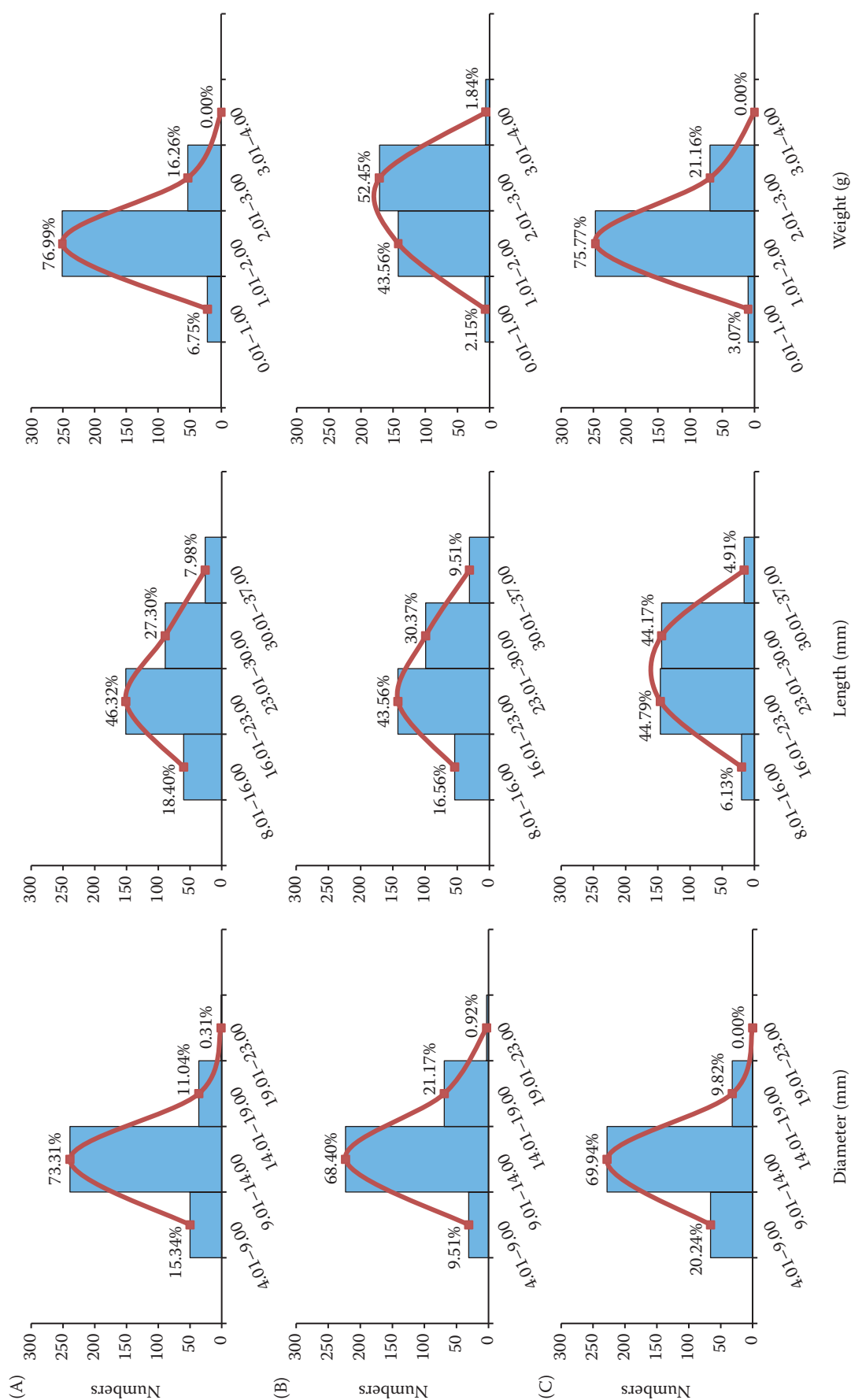


Figure 2. Effects of washing and blanching on the dimensions and weights of shiitake stipes: (A) fresh stipes, (B) washed stipes and (C) blanched stipes ( $n = 326$ )



Table 1. Proximate compositions of fresh, washed and blanched shiitake (%; mean  $\pm$  SD;  $n = 3$ )

|              | Cap content                  |                              |                               | Stipe content                 |                               |                                |
|--------------|------------------------------|------------------------------|-------------------------------|-------------------------------|-------------------------------|--------------------------------|
|              | fresh                        | washed                       | blanched                      | fresh                         | washed                        | blanched                       |
| Moisture*    | 92.9 $\pm$ 0.2 <sup>Ab</sup> | 94.6 $\pm$ 0.5 <sup>Aa</sup> | 94.2 $\pm$ 0.2 <sup>Aa</sup>  | 91.2 $\pm$ <0.1 <sup>Ac</sup> | 92.7 $\pm$ 0.2 <sup>Ab</sup>  | 92.0 $\pm$ 1.0 <sup>Abc</sup>  |
| Protein      | 18.5 $\pm$ 0.2 <sup>Ba</sup> | 18.4 $\pm$ 0.3 <sup>Ba</sup> | 16.3 $\pm$ 0.9 <sup>Bb</sup>  | 15.5 $\pm$ 0.4 <sup>Bbc</sup> | 15.5 $\pm$ 0.3 <sup>Bbc</sup> | 14.6 $\pm$ < 0.1 <sup>Bc</sup> |
| Fat          | 3.0 $\pm$ 0.2 <sup>Da</sup>  | 3.0 $\pm$ 0.2 <sup>Da</sup>  | 2.6 $\pm$ < 0.1 <sup>Cb</sup> | 2.6 $\pm$ 0.1 <sup>Db</sup>   | 2.6 $\pm$ 0.1 <sup>Db</sup>   | 2.1 $\pm$ 0.1 <sup>Dc</sup>    |
| Ash          | 6.2 $\pm$ 0.2 <sup>Ca</sup>  | 6.3 $\pm$ 0.3 <sup>Ca</sup>  | 3.7 $\pm$ 0.2 <sup>Cc</sup>   | 4.3 $\pm$ 0.1 <sup>Cb</sup>   | 4.2 $\pm$ 0.1 <sup>Cb</sup>   | 3.4 $\pm$ 0.2 <sup>Cc</sup>    |
| Carbohydrate | 72.3 $\pm$ 0.3 <sup>Ac</sup> | 72.3 $\pm$ 0.5 <sup>Ac</sup> | 77.4 $\pm$ 0.7 <sup>Ab</sup>  | 77.6 $\pm$ 0.5 <sup>Ab</sup>  | 77.7 $\pm$ 0.3 <sup>Ab</sup>  | 79.9 $\pm$ 0.1 <sup>Aa</sup>   |

\*Moisture was presented on air-dried matter basis, others were presented on a dry matter basis; <sup>A–D</sup>values with different capital letters within a column differ significantly ( $P < 0.05$ ); <sup>a–c</sup>values with different lowercase letters within a row differ significantly ( $P < 0.05$ ); SD – standard deviation

Table 2. Mineral components of fresh, washed and blanched shiitake (mg kg<sup>−1</sup> dry matter; mean  $\pm$  SD;  $n = 3$ )

|                      | Cap content                      |                                  |                                 | Stipe content                    |                                  |                                 |
|----------------------|----------------------------------|----------------------------------|---------------------------------|----------------------------------|----------------------------------|---------------------------------|
|                      | fresh                            | washed                           | blanched                        | fresh                            | washed                           | blanched                        |
| <b>Major mineral</b> |                                  |                                  |                                 |                                  |                                  |                                 |
| Ca                   | 141.0 $\pm$ 5 <sup>Cb</sup>      | 143.0 $\pm$ 6 <sup>Cb</sup>      | 79.7 $\pm$ 3.3 <sup>Cd</sup>    | 158.0 $\pm$ 6 <sup>Ca</sup>      | 163.0 $\pm$ 8 <sup>Ca</sup>      | 120.0 $\pm$ 5 <sup>Cc</sup>     |
| Mg                   | 918.0 $\pm$ 5 <sup>Ba</sup>      | 912.0 $\pm$ 8 <sup>Ba</sup>      | 521.0 $\pm$ 4 <sup>Bd</sup>     | 788.0 $\pm$ 31 <sup>Bb</sup>     | 780.0 $\pm$ 43 <sup>Bb</sup>     | 602.0 $\pm$ 27 <sup>Bc</sup>    |
| P                    | 12.5 $\pm$ 0.3 <sup>Ea</sup>     | 13.3 $\pm$ 0.9 <sup>Ea</sup>     | 7.7 $\pm$ 0.3 <sup>Ec</sup>     | 9.2 $\pm$ 0.4 <sup>Cb</sup>      | 9.0 $\pm$ 0.5 <sup>Cb</sup>      | 7.0 $\pm$ 0.3 <sup>Cc</sup>     |
| K                    | 15 325.0 $\pm$ 123 <sup>Aa</sup> | 15 407.0 $\pm$ 168 <sup>Aa</sup> | 8 505.0 $\pm$ 69 <sup>Ac</sup>  | 12 177.0 $\pm$ 456 <sup>Ab</sup> | 12 209.0 $\pm$ 502 <sup>Ab</sup> | 9 390.0 $\pm$ 353 <sup>Ac</sup> |
| Na                   | 89.0 $\pm$ 4.4 <sup>Da</sup>     | 84.3 $\pm$ 5.2 <sup>Da</sup>     | 51.5 $\pm$ 2.9 <sup>Dc</sup>    | 49.8 $\pm$ 2.5 <sup>Cb</sup>     | 50.0 $\pm$ 4.6 <sup>Cb</sup>     | 39.6 $\pm$ 2.3 <sup>Cd</sup>    |
| <b>Trace mineral</b> |                                  |                                  |                                 |                                  |                                  |                                 |
| Al                   | 1.07 $\pm$ 0.02 <sup>Eb</sup>    | 1.04 $\pm$ 0.09 <sup>Eb</sup>    | 0.61 $\pm$ 0.03 <sup>Ec</sup>   | 1.50 $\pm$ 0.06 <sup>Ca</sup>    | 1.44 $\pm$ 0.11 <sup>Ca</sup>    | 1.14 $\pm$ 0.06 <sup>Cb</sup>   |
| Cr                   | 0.21 $\pm$ 0.01 <sup>Eb</sup>    | 0.22 $\pm$ 0.01 <sup>Eb</sup>    | 0.12 $\pm$ 0.02 <sup>Ed</sup>   | 0.25 $\pm$ 0.02 <sup>Ca</sup>    | 0.24 $\pm$ 0.03 <sup>Ca</sup>    | 0.19 $\pm$ 0.01 <sup>Cc</sup>   |
| Fe                   | 7.87 $\pm$ 0.19 <sup>Ea</sup>    | 7.71 $\pm$ 0.24 <sup>Ea</sup>    | 4.50 $\pm$ 0.11 <sup>Ec</sup>   | 7.77 $\pm$ 0.37 <sup>Ca</sup>    | 8.01 $\pm$ 0.52 <sup>Ca</sup>    | 6.06 $\pm$ 0.36 <sup>Cb</sup>   |
| Mn                   | 7.77 $\pm$ 0.26 <sup>Eb</sup>    | 7.87 $\pm$ 0.42 <sup>Eb</sup>    | 4.53 $\pm$ 0.18 <sup>Ed</sup>   | 8.38 $\pm$ 0.41 <sup>Ca</sup>    | 8.02 $\pm$ 0.51 <sup>Ca</sup>    | 6.11 $\pm$ 0.31 <sup>Cc</sup>   |
| Se                   | 6.08 $\pm$ 0.24 <sup>Eb</sup>    | 5.99 $\pm$ 0.37 <sup>Eb</sup>    | 3.40 $\pm$ 0.17 <sup>Ed</sup>   | 7.14 $\pm$ 0.36 <sup>Ca</sup>    | 6.92 $\pm$ 0.49 <sup>Ca</sup>    | 5.41 $\pm$ 0.42 <sup>Cc</sup>   |
| Zn                   | 66.70 $\pm$ 3.2 <sup>Da</sup>    | 67.20 $\pm$ 4.4 <sup>Da</sup>    | 37.50 $\pm$ 2.4 <sup>Dd</sup>   | 56.00 $\pm$ 2.3 <sup>Cb</sup>    | 58.70 $\pm$ 4.2 <sup>Cb</sup>    | 44.00 $\pm$ 2.9 <sup>Cc</sup>   |
| <b>Heavy metal</b>   |                                  |                                  |                                 |                                  |                                  |                                 |
| Cd                   | 0.20 $\pm$ 0.01 <sup>Ea</sup>    | 0.19 $\pm$ < 0.01 <sup>Ea</sup>  | 0.12 $\pm$ < 0.01 <sup>Ec</sup> | 0.20 $\pm$ 0.01 <sup>Ca</sup>    | 0.20 $\pm$ 0.01 <sup>Ca</sup>    | 0.16 $\pm$ 0.01 <sup>Cb</sup>   |
| Pb                   | 0.05 $\pm$ < 0.01 <sup>Ea</sup>  | 0.05 $\pm$ < 0.01 <sup>Ea</sup>  | 0.03 $\pm$ < 0.01 <sup>Eb</sup> | 0.05 $\pm$ < 0.01 <sup>Ca</sup>  | 0.06 $\pm$ 0.01 <sup>Ca</sup>    | 0.04 $\pm$ 0.01 <sup>Cb</sup>   |

<sup>A–E</sup>values with different capital letters within a column differ significantly ( $P < 0.05$ ); <sup>a–d</sup>values with different lowercase letters within a row differ significantly ( $P < 0.05$ ); SD – standard deviation

Table 3. Sugars and polyols of fresh, washed and blanched shiitake (mg g<sup>−1</sup> dry matter; mean  $\pm$  SD;  $n = 3$ )

|           | Cap content                  |                              |                              | Stipe content                |                              |                              |
|-----------|------------------------------|------------------------------|------------------------------|------------------------------|------------------------------|------------------------------|
|           | fresh                        | washed                       | blanched                     | fresh                        | washed                       | blanched                     |
| Trehalose | 19.0 $\pm$ 0.4 <sup>Ba</sup> | 12.3 $\pm$ 0.3 <sup>Bb</sup> | 5.2 $\pm$ 0.05 <sup>Bd</sup> | 10.3 $\pm$ 0.5 <sup>Cc</sup> | 4.0 $\pm$ 0.1 <sup>Ce</sup>  | 3.6 $\pm$ 0.1 <sup>Ce</sup>  |
| Arabitrol | 8.8 $\pm$ 0.1 <sup>Cd</sup>  | 6.8 $\pm$ 0.3 <sup>Ce</sup>  | 4.0 $\pm$ 0.07 <sup>Bf</sup> | 63.6 $\pm$ 1.8 <sup>Ba</sup> | 51.5 $\pm$ 1.1 <sup>Bb</sup> | 27.1 $\pm$ 1.4 <sup>Bc</sup> |
| Mannitol  | 115 $\pm$ 3 <sup>Aab</sup>   | 118 $\pm$ 2 <sup>Aa</sup>    | 80 $\pm$ 1 <sup>Ac</sup>     | 96 $\pm$ 4 <sup>Ab</sup>     | 98 $\pm$ 4 <sup>Ab</sup>     | 72 $\pm$ 2 <sup>Ad</sup>     |

<sup>A–C</sup>values with different capital letters within a column differ significantly ( $P < 0.05$ ); <sup>a–f</sup>values with different lowercase letters within a row differ significantly ( $P < 0.05$ ); SD – standard deviation

Li et al. (2017) (18.73 mg g<sup>−1</sup>), but higher than those of Yang et al. (2001) (1.71–1.93 mg g<sup>−1</sup>).

**5'-Nucleotides.** Among the six 5'-nucleotides of tested samples (Table 5), 5'-adenosine monophosphate (5'-AMP), 5'-guanosine monophosphate (5'-GMP), 5'-ino-

sine monophosphate (5'-IMP), and 5'-xanthosine monophosphate (5'-XMP) have umami flavour (Dermiki et al. 2013). In particular, 5'-GMP has a meaty taste and is a flavour enhancer that is more effective than MSG (Yang et al. 2001). The 5'-nucleotide concentrations

Table 4. Free amino acids (AA) of fresh, washed and blanched shiitake (mg g<sup>-1</sup> dry matter; mean ± SD; *n* = 3)

|                                 | Cap content                |                             |                             | Stipe content              |                             |                             |
|---------------------------------|----------------------------|-----------------------------|-----------------------------|----------------------------|-----------------------------|-----------------------------|
|                                 | fresh                      | washed                      | blanched                    | fresh                      | washed                      | blanched                    |
| <b>Essential amino acid</b>     |                            |                             |                             |                            |                             |                             |
| Histidine                       | 0.61 ± 0.06 <sup>LMa</sup> | 0.61 ± 0.06 <sup>Ja</sup>   | 0.32 ± 0.02 <sup>LMb</sup>  | 0.45 ± 0.02 <sup>Jb</sup>  | 0.44 ± 0.04 <sup>Kb</sup>   | 0.22 ± 0.012 <sup>Od</sup>  |
| Isoleucine                      | 0.44 ± 0.03 <sup>Nc</sup>  | 0.44 ± 0.04 <sup>Kc</sup>   | 0.36 ± 0.01 <sup>KLd</sup>  | 1.00 ± 0.03 <sup>Hla</sup> | 1.00 ± 0.02 <sup>IJa</sup>  | 0.50 ± 0.02 <sup>Lb</sup>   |
| Leucine                         | 0.64 ± 0.01 <sup>Lc</sup>  | 0.62 ± 0.03 <sup>Jc</sup>   | 0.43 ± < 0.01 <sup>Jd</sup> | 1.54 ± 0.07 <sup>FGa</sup> | 1.47 ± 0.03 <sup>Hb</sup>   | 0.65 ± 0.02 <sup>JKc</sup>  |
| Lysine                          | 1.77 ± 0.16 <sup>Hab</sup> | 1.88 ± 0.12 <sup>Fa</sup>   | 1.53 ± 0.02 <sup>Ecd</sup>  | 1.68 ± 0.01 <sup>Fbc</sup> | 1.66 ± 0.15 <sup>Gbc</sup>  | 1.41 ± 0.07 <sup>Ed</sup>   |
| Methionine                      | 5.14 ± 0.09 <sup>Ba</sup>  | 5.18 ± 0.06 <sup>Aa</sup>   | 3.27 ± 0.12 <sup>Ab</sup>   | nd                         | nd                          | nd                          |
| Phenylalanine                   | 0.49 ± 0.05 <sup>MNb</sup> | 0.52 ± 0.02 <sup>JKb</sup>  | 0.37 ± 0.04 <sup>KLc</sup>  | 0.83 ± 0.03 <sup>Ia</sup>  | 0.85 ± 0.01 <sup>Ia</sup>   | 0.42 ± 0.01 <sup>Mc</sup>   |
| Threonine                       | 2.47 ± 0.08 <sup>Fa</sup>  | 2.42 ± 0.13 <sup>Eab</sup>  | 1.50 ± 0.03 <sup>Ed</sup>   | 2.21 ± 0.22 <sup>Ec</sup>  | 2.23 ± 0.01 <sup>Fbc</sup>  | 1.34 ± < 0.01 <sup>Fd</sup> |
| Tryptophan                      | 0.27 ± 0.03 <sup>Oc</sup>  | 0.25 ± 0.04 <sup>Lc</sup>   | 0.19 ± 0.03 <sup>Nc</sup>   | 3.48 ± 0.27 <sup>Ba</sup>  | 3.39 ± 0.31 <sup>Ca</sup>   | 2.74 ± 0.05 <sup>Bb</sup>   |
| Valine                          | 2.06 ± 0.04 <sup>Ga</sup>  | 2.05 ± 0.15 <sup>Fa</sup>   | 1.07 ± 0.02 <sup>Gb</sup>   | 2.04 ± 0.07 <sup>Ea</sup>  | 2.06 ± 0.02 <sup>Fa</sup>   | 0.94 ± 0.02 <sup>Hb</sup>   |
| <b>Non-essential amino acid</b> |                            |                             |                             |                            |                             |                             |
| Alanine                         | 3.55 ± 0.15 <sup>Eab</sup> | 3.49 ± 0.21 <sup>Db</sup>   | 1.23 ± 0.01 <sup>Fc</sup>   | 3.65 ± 0.08 <sup>Bab</sup> | 3.75 ± 0.05 <sup>Ba</sup>   | 1.14 ± 0.02 <sup>Gc</sup>   |
| Arginine                        | 4.09 ± 0.11 <sup>Da</sup>  | 4.01 ± 0.20 <sup>Ca</sup>   | 2.93 ± < 0.01 <sup>Cb</sup> | 3.09 ± 0.20 <sup>Cb</sup>  | 3.07 ± 0.04 <sup>Db</sup>   | 1.71 ± 0.03 <sup>Dc</sup>   |
| Asparagine                      | 1.40 ± 0.06 <sup>IJa</sup> | 1.36 ± 0.09 <sup>GHa</sup>  | 0.75 ± 0.03 <sup>Hc</sup>   | 1.15 ± 0.05 <sup>Hb</sup>  | 1.13 ± < 0.01 <sup>Ib</sup> | 0.61 ± 0.01 <sup>Kd</sup>   |
| Aspartic acid                   | 0.66 ± 0.01 <sup>La</sup>  | 0.64 ± 0.03 <sup>Ja</sup>   | 0.40 ± 0.01 <sup>JKb</sup>  | 0.42 ± 0.03 <sup>Jb</sup>  | 0.40 ± 0.01 <sup>Kb</sup>   | 0.29 ± 0.01 <sup>INc</sup>  |
| Cystine                         | nd                         | nd                          | nd                          | nd                         | nd                          | nd                          |
| Glutamic acid                   | 4.83 ± 0.08 <sup>Ca</sup>  | 4.86 ± 0.08 <sup>Ba</sup>   | 2.47 ± 0.03 <sup>Db</sup>   | 2.50 ± 0.09 <sup>Db</sup>  | 2.42 ± 0.17 <sup>Eb</sup>   | 2.09 ± 0.04 <sup>Cc</sup>   |
| Glutamine                       | 5.30 ± 0.13 <sup>Ab</sup>  | 5.14 ± 0.28 <sup>Ab</sup>   | 3.09 ± 0.03 <sup>Bd</sup>   | 8.69 ± 0.14 <sup>Aa</sup>  | 8.68 ± 0.13 <sup>Aa</sup>   | 4.59 ± 0.09 <sup>Ac</sup>   |
| Glycine                         | 1.33 ± 0.02 <sup>Jb</sup>  | 1.27 ± 0.07 <sup>Hb</sup>   | 0.67 ± 0.01 <sup>Lc</sup>   | 1.70 ± 0.01 <sup>Fa</sup>  | 1.72 ± 0.11 <sup>Ga</sup>   | 0.70 ± 0.03 <sup>IJc</sup>  |
| Proline                         | 0.58 ± 0.01 <sup>LMa</sup> | 0.58 ± < 0.01 <sup>Ja</sup> | 0.28 ± < 0.01 <sup>Md</sup> | 0.42 ± 0.01 <sup>Jb</sup>  | 0.42 ± < 0.01 <sup>Kb</sup> | 0.34 ± < 0.01 <sup>Nc</sup> |
| Serine                          | 1.46 ± 0.04 <sup>Ia</sup>  | 1.42 ± 0.08 <sup>Ga</sup>   | 0.80 ± < 0.01 <sup>Hb</sup> | 1.42 ± 0.12 <sup>Ga</sup>  | 1.41 ± 0.03 <sup>Ha</sup>   | 0.73 ± < 0.01 <sup>Ib</sup> |
| Tyrosine                        | 0.80 ± 0.07 <sup>Ka</sup>  | 0.79 ± 0.05 <sup>Ia</sup>   | 0.43 ± 0.02 <sup>Kb</sup>   | nd                         | nd                          | nd                          |

A–O values with different capital letters within a column differ significantly ( $P < 0.05$ ); a–d values with different lowercase letters within a row differ significantly ( $P < 0.05$ ); SD – standard deviation; nd – not detected

in fresh caps and stipes was in the descending order of 5'-AMP ≈ 5'-cytidine monophosphate (5'-CMP) ≈ 5'-GMP > 5'-uridine monophosphate (5'-UMP) > 5'-XMP ≈ 5'-IMP. The contents of each flavour and total 5'-nucleotides in the caps were significantly higher than those in the stipes, which is similar to the results of Chen et al. (2015) and Li et al. (2018). Blanching showed a significant decrease of 5'-nucleotide contents in shiitake, which was similar to the results of Li et al. (2017).

**EUC.** Dermiki et al. (2013) reported that shiitake is one of the foods with rich umami taste, which is composed of aspartic acid, glutamic acid, 5'-GMP, 5'-XMP, 5'-IMP, and 5'-AMP. The EUC (g MSG 100 g<sup>-1</sup>) of FC and WC are 315 and 310, respectively, which means that the concentration of umami is about 3-fold that of MSG, and the EUC of fresh shiitake was not affected by washing. However, the EUC of BC and BS dropped to 30.4 and 56.3, respectively, which means that the blanched broth contains lots of EUC. The effect of blanching on the EUC of shiitake was similar

to that of Li et al. (2017). The EUC of FC and FS values in this study were higher than those of Chen et al. (2015) (at maturity stage, about 140–150 and 35–75, respectively), but lower than the results (728.54 and 148.99, respectively) of Li et al. (2018). This result might be due to different varieties of mushrooms and cultivation conditions.

**Bioactive compounds.** Ergothioneine is related to autoimmune disorders and excellent antioxidant *in vivo*, but it is only produced by fungi and some prokaryotes (Cohen et al. 2014). The ergothioneine content of samples was in the descending order of FC ≈ WC > FS ≈ WS > BC ≈ BS (Table 6). Since ergothioneine is water-soluble, hot water blanching caused the loss of ergothioneine. Some food products containing GABA had shown protection and hypotensive actions on hypertensive patients (Tanaka et al. 2009). The GABA content of samples was in the order of FC ≈ WC > FS ≈ WS > BC ≈ BS. Compared to published results (Chen et al. 2012; Cohen et al. 2014), the GABA content in FC and FS was found

Table 5. 5'-Nucleotides and equivalent umami concentration (EUC) of fresh, washed and blanched shiitake (mg g<sup>-1</sup> dry matter; mean ± SD; *n* = 3)

|                                           | Cap content               |                            |                           | Stipe content              |                           |                             |
|-------------------------------------------|---------------------------|----------------------------|---------------------------|----------------------------|---------------------------|-----------------------------|
|                                           | fresh                     | washed                     | blanched                  | fresh                      | washed                    | blanched                    |
| 5'-AMP                                    | 1.93 ± 0.13 <sup>Aa</sup> | 1.85 ± 0.02 <sup>Aa</sup>  | 0.58 ± 0.01 <sup>Bd</sup> | 1.36 ± 0.14 <sup>Ab</sup>  | 1.36 ± 0.03 <sup>Ab</sup> | 0.80 ± 0.05 <sup>Ac</sup>   |
| 5'-CMP                                    | 1.78 ± 0.03 <sup>Aa</sup> | 1.74 ± 0.05 <sup>ABa</sup> | 1.21 ± 0.01 <sup>Ab</sup> | 1.18 ± 0.04 <sup>Abc</sup> | 1.16 ± 0.06 <sup>Bc</sup> | 0.62 ± 0.06 <sup>Cd</sup>   |
| 5'-GMP                                    | 1.76 ± 0.24 <sup>Aa</sup> | 1.71 ± 0.03 <sup>Ba</sup>  | 0.13 ± 0.01 <sup>Fd</sup> | 1.14 ± 0.15 <sup>Ab</sup>  | 1.09 ± 0.03 <sup>Bb</sup> | 0.76 ± 0.09 <sup>ABc</sup>  |
| 5'-IMP                                    | 0.53 ± 0.05 <sup>Ca</sup> | 0.56 ± 0.06 <sup>Da</sup>  | 0.44 ± 0.01 <sup>Cb</sup> | 0.31 ± 0.01 <sup>Cc</sup>  | 0.32 ± 0.01 <sup>Ec</sup> | 0.18 ± < 0.01 <sup>Dd</sup> |
| 5'-UMP                                    | 1.56 ± 0.08 <sup>Ba</sup> | 1.54 ± 0.08 <sup>Ca</sup>  | 0.28 ± 0.02 <sup>Dc</sup> | 0.73 ± 0.19 <sup>Bb</sup>  | 0.75 ± 0.10 <sup>Cb</sup> | 0.68 ± 0.01 <sup>BCb</sup>  |
| 5'-XMP                                    | 0.60 ± 0.01 <sup>Ca</sup> | 0.59 ± 0.09 <sup>Da</sup>  | 0.23 ± 0.01 <sup>Ed</sup> | 0.42 ± 0.02 <sup>Cb</sup>  | 0.47 ± 0.06 <sup>Db</sup> | 0.16 ± 0.03 <sup>Dc</sup>   |
| EUC (g MSG<br>100 g <sup>-1</sup> sample) | 315 ± 36 <sup>a</sup>     | 310 ± <1 <sup>a</sup>      | 30.4 ± 0.7 <sup>c</sup>   | 106 ± 6 <sup>b</sup>       | 100 ± 7 <sup>b</sup>      | 56.3 ± 4.2 <sup>c</sup>     |

<sup>A–F</sup>values with different capital letters within a column differ significantly (*P* < 0.05); <sup>a–d</sup>values with different lowercase letters within a row differ significantly (*P* < 0.05); SD – standard deviation; 5'-AMP – 5'-adenosine monophosphate; 5'-CMP – 5'-cytidine monophosphate; 5'-GMP – 5'-guanosine monophosphate; 5'-IMP – 5'-inosine monophosphate; 5'-UMP – 5'-uridine monophosphate; 5'-XMP – 5'-xanthosine monophosphate; MSG – monosodium glutamate

Table 6. Bioactive compounds of fresh, washed and blanched shiitake (mg g<sup>-1</sup> dry matter; mean ± SD; *n* = 3)

|                                                      | Cap content              |                          |                           | Stipe content            |                           |                           |
|------------------------------------------------------|--------------------------|--------------------------|---------------------------|--------------------------|---------------------------|---------------------------|
|                                                      | fresh                    | washed                   | blanched                  | fresh                    | washed                    | blanched                  |
| Ergothioneine                                        | 1.02 ± 0.06 <sup>a</sup> | 0.97 ± 0.01 <sup>a</sup> | 0.44 ± 0.03 <sup>cd</sup> | 0.56 ± 0.01 <sup>b</sup> | 0.51 ± 0.04 <sup>bc</sup> | 0.41 ± <0.01 <sup>d</sup> |
| γ-aminobutyric acid                                  | 4.95 ± 0.17 <sup>a</sup> | 4.91 ± 0.31 <sup>a</sup> | 1.74 ± 0.02 <sup>c</sup>  | 2.14 ± 0.20 <sup>b</sup> | 2.10 ± 0.13 <sup>b</sup>  | 1.54 ± 0.09 <sup>c</sup>  |
| Ergosterol                                           | 4.08 ± 0.02 <sup>a</sup> | 3.95 ± 0.05 <sup>a</sup> | 3.59 ± 0.04 <sup>b</sup>  | 3.68 ± 0.05 <sup>b</sup> | 3.63 ± 0.06 <sup>b</sup>  | 3.36 ± 0.05 <sup>c</sup>  |
| Total phenols<br>(mg GAE g <sup>-1</sup> dry matter) | 1.76 ± 0.05 <sup>a</sup> | 1.67 ± 0.01 <sup>a</sup> | 0.93 ± 0.03 <sup>cd</sup> | 1.25 ± 0.06 <sup>b</sup> | 1.08 ± 0.05 <sup>bc</sup> | 0.81 ± 0.02 <sup>d</sup>  |
| Crude polysaccharide                                 | 38.3 ± 0.5 <sup>b</sup>  | 37.0 ± 0.2 <sup>b</sup>  | 30.9 ± 0.51 <sup>d</sup>  | 40.4 ± 0.42 <sup>a</sup> | 39.9 ± 0.28 <sup>a</sup>  | 33.5 ± 0.41 <sup>c</sup>  |

<sup>a–d</sup>Values with different lowercase letters within a row differ significantly (*P* < 0.05); SD – standard deviation; GAE – gallic acid equivalent

to be lower. The caps had significantly higher ergosterol and total phenols content than those in the stipes. Contrary to other bioactive components, crude polysaccharide content was significantly higher in the stipes than in the caps.

## CONCLUSION

According to the results of this study, washing and blanching affected the dimensions and weights of FC and FS. On a dry basis, washing mainly affected the sugars and polyols content in all tested samples, but other nutrients and bioactive compounds were not affected. Blanching significantly affected the nutrients and bioactive compounds of all tested samples. Moisture, protein, fat, ash, potassium, magnesium, mannitol, trehalose, MSG-like and bitter AA, 5'-nucleotides, EUC, ergosterol, ergothioneine, GABA, and total phenols contents in the stipes were significantly lower than those in the caps, while carbohydrate, calcium, arabinol,

sweet AA, and crude polysaccharides contents in the stipes were higher than those in the caps. These results indicate that the stipes should not be underestimated or discarded because it contains the necessary nutrients and bioactive compounds, and it should be considered to develop it into new products to improve its economic efficiency. Besides, from the nutrient content of the caps and stipes after blanching, it can be speculated that the blanched broth may contain a large amount of nutrients and bioactive compounds, which should be applied and developed into new products. Related research on the development of blanched broth into an instant drink is in progress.

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Received: September 1, 2020

Accepted: August 23, 2021