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# **Nonvolatile Taste Components of Ear Mushrooms**

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Five kinds of ear mushrooms are commercially available in Taiwan, including black, red, jin, snow, and silver ears. Ash contents ranged from 2.05 to 6.14% of dry weight. Carbohydrate contents ranged from 68.88 to 88.14%. The jin and snow ears contained high amounts of crude fat (4.48 and 4.54%, respectively). The jin and snow ears also contained high amounts of crude fiber (11.69 and 8.51%, respectively). Crude protein contents were high in the black and snow ears (15.46 and 12.53%, respectively). Ear mushrooms contained very low amounts of soluble sugars but high amounts of other reducing sugars. Total free amino acid contents ranged from 0.53 to 1.24 mg/g. Monosodium glutamate-like component contents ranged from 0.05 to 0.34 mg/g. Sweet component contents ranged from 0.17 to 0.50 mg/g. Bitter component content was high in the silver ear. Total 5'-nucleotides contents ranged from 0.69 to 5.39 mg/g. Flavor 5'-nucleotide contents were high in the snow and jin ears. In this study, the five kinds were considerably different in both their proximate compositions and taste components.

**Keywords:** Ear mushrooms; Auricularia fuscosuccinea; Auricularia mesenteria; Auricularia polytricha; Tremella fuciformis; soluble sugars; free amino acids; 5'-nucleotides

#### INTRODUCTION

Ear mushrooms are peculiar mushrooms that have captured the palate of Asian mycophagists for centuries. Currently, five kinds of ear mushrooms are commercially available in Taiwan, including black, red, jin, snow, and silver ears (Wu, 1995). The black ear [Auricularia mesenteria (Dickson) Persoon] is dark brown and velvety on the upper surface and pinkish gray on the lower surface. The red ear [A. polytricha (Montagne) Saccardol is brownish and finely hairy on its outer surface. The jin ear [A. fuscosuccinea (Montagne) Farlow, brown strain is light brown, very thin, and hairless with a more rubbery texture. The snow ear [A. fuscosuccinea (Montagne) Farlow, white strain], which is a newly cultivated edible ear mushroom in Taiwan, is a stable translucent white mutant of jin ears. The silver ear (Tremella fuciformis Berkeley) is a translucent white species of jelly fungi. In general, the cultivation methods for these ear mushrooms are parallel to that of shiitake mushrooms (Lentinula edodes) on logs or on sterilized sawdust (Tu and Wu, 1989).

These ear mushrooms rehydrate readily from a dried state, embellish soups and sauces, and impart a unique and pleasing texture to most meals (Tu and Wu, 1989). Therefore, these ear mushrooms are highly valued as a centerpiece of Asian cooking. However, the taste components of these five ear mushrooms are unknown. Our objective was to examine the nonvolatile taste components in the five ear mushrooms, including their proxi-

mate compositions, soluble sugars, free amino acids, and 5'-nucleotides.

#### MATERIALS AND METHODS

**Mushrooms.** Fresh jin and snow ears were harvested from the mushroom farm of the Taiwan Agricultural Research Institute, Taichung County, Taiwan. Fresh black ears were obtained from Taichung County, Taiwan. Fresh ear mushrooms from each kind were randomly selected into three samples,  $\sim\!500$  g each. Fresh ear mushrooms were air-dried in an oven at 60 °C before analysis. Dried red and silver ears were purchased at a local market in Taichung City, Taiwan. Dried ear mushrooms were also randomly selected into three samples,  $\sim\!50$  g each.

**Proximate Analysis.** The proximate compositions of five ear mushrooms, including moisture, ash, carbohydrate, crude fat, crude fiber, and crude protein, were determined according to the methods of the AOAC (1990). The nitrogen factor used for crude protein calculation was 4.38 (Crisan and Sands, 1978).

**Soluble Sugar Assay.** Soluble sugars were extracted and analyzed as described by Ajlouni et al. (1995). Air-dried ear powder (600 mg) was extracted with 50 mL of 80% aqueous ethanol (95% pure, Taiwan Tobacco and Wine Monopoly Bureau, Taipei), and xylose (50 mg, Sigma Chemical Co., St. Louis, MO) was added as an internal standard. This suspension was shaken for 45 min at room temperature and filtered through Whatman No. 4 filter paper. The residue was washed five times with additional 25-mL portions of 80% ethanol. The combined filtrate was then rotary evaporated at 40 °C and redissolved in deionized water to a final volume of 10 mL. The aqueous extract was passed through a filter unit (13 mm, Lida Corp., Kenosha, WI) and filtered using a 0.45-μm CA nonsterile filter (Lida) prior to injection onto a high-performance liquid chromatograph (HPLC).

The HPLC system consisted of a Hitachi L-6000 pump, a Rheodyne 7161 injector, a 20- $\mu$ L sample loop, a Hitachi D-2500 chromatointegrator, a Bischoff RI 8110 detector, and a Phase Sep-NH<sub>2</sub> column ( $4.6 \times 250$  mm,  $5\,\mu$ m, Phase Separation Inc.,

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Norwalk, CT). The mobile phase was acetonitrile (LC grade, Tedia Co., Fairfield, OH)/deionized water, 90:10 (v/v), at a flow rate of 1 mL/min. Each sugar was quantified by comparing the peak area of the sugar to that of the internal standard.

**Total Reducing Sugar Determination.** Reducing sugars were determined using the 3,5-dinitrosalicylic acid method as described by James (1995). The absorbance of each sample solution was measured at 540 nm on a Hitachi 100-60 spectrophotometer. Total reducing sugars were calculated on the basis of the calibration curve of glucose.

**Free Amino Acid Assay.** Freeze-dried ear powder (500 mg) was shaken with 50 mL of 0.1 N HCl (Union Chemical Co., Hsinchu, Taiwan) for 45 min at ambient temperature and filtered through Whatman No. 4 filter paper. The filtrate was then passed through a filter unit (13 mm, Lida), and filtered using a 0.45- $\mu$ m CA nonsterile filter (Lida). The purified filtrate was mixed with o-phthalaldehyde reagent (Sigma) in an Eppendorf tube, shaken to facilitate derivatization, and then immediately injected onto the HPLC.

The HPLC system was the same as for sugar analysis but included a Hitachi F-1050 fluorescence detector, with fluorescence excitation at 340 nm and emission at 450 nm, and a Prodigy 5 ODS-2 column (4.6  $\times$  250 mm, 5  $\mu$ m, Phenomenex Inc., Torrance, CA). The mobile phases and gradient conditions were the same as described in Mau et al. (1997). Each amino acid was quantified by the calibration curve of the authentic amino acid.

5'-Nucleotide Assay. 5'-Nucleotides were extracted and analyzed as described by Taylor et al. (1981). Freeze-dried ear powder (500 mg) was extracted with 25 mL of deionized water. This suspension was heated to boiling for 1 min, cooled, and then centrifuged at 22200g for 15 min. The extraction was repeated once with 20 mL of deionized water. The combined filtrate was then evaporated and filtered prior to HPLC injection in the same manner as in the soluble sugar assay.

The HPLC system was the same as for the sugar assay except for a Hitachi L-4000 UV detector and a Prodigy 5 ODS-2 column (4.6  $\times$  250 mm, 5  $\mu m$ , Phenomenex). The mobile phase was 0.5 M KH<sub>2</sub>PO<sub>4</sub>/H<sub>3</sub>PO<sub>4</sub> (pH 4.0, Wako Pure Chemical Co., Osaka, Japan) at a flow rate of 1 mL/min and UV detection at 254 nm. Each 5′-nucleotide was quantified by the calibration curve of the authentic 5′-nucleotide.

**Statistical Analysis.** For each ear mushroom, three samples were used for the determination of every quality attribute. The experimental data were subjected to an analysis of variance for a completely random design as described by Steel et al. (1997) to determine the least significant difference among means at the level of 0.05. After multiple comparisons, the means in the following tables were followed with different capital letters A—D on the basis of their values and statistical differences. In the case that a mean was followed with AB, this mean was not significantly different from a mean with A and was not significantly different from another mean with B. However, means with different letters were significantly different at the level of 0.05.

# RESULTS AND DISCUSSION

As commercial dried products, the jin ear is more popular and competes with the black and red ears, whereas the snow ear competes with the silver ear. The moisture contents varied among dried ear mushrooms and ranged from 10.81 to 12.31% (Table 1). The ash contents in the five ear mushrooms ranged from 2.05 to 6.14% and were in the order of the red, black, jin, snow, and silver ears. The carbohydrate contents ranged from 68.88 to 88.14% and were in the order of the snow, jin, black, silver, and red ears. Two strains of *A. fuscosuccinea* (the jin and snow ears) contained the highest amount of crude fat (4.48 and 4.54% of dry weight, respectively). However, the jin ear contained the highest amount of crude fiber (11.69%), and the

**Table 1. Proximate Composition in Ear Mushrooms** 

	content <sup>b</sup> (%)				
component <sup>a</sup>	black ear <sup>c</sup>	$\operatorname{red}_{\operatorname{ear}^c}$	$ \frac{\text{jin}}{\text{ear}^c} $	snow ear <sup>c</sup>	silver ear <sup>c</sup>
moisture	11.69C	12.05B	12.31A	10.81D	11.64C
dry matter	88.31B	87.95C	87.69D	89.19A	88.36B
ash	3.29D	2.05E	4.02C	5.54B	6.14A
carbohydrate	76.53C	88.14A	71.19D	68.88D	81.72B
crude fat	0.80B	0.48C	4.48A	4.54A	0.93B
crude fiber	3.92C	3.63C	11.69A	8.51B	2.91C
crude protein	15.46A	5.70D	8.62C	12.53B	8.30C

<sup>a</sup> Moisture and dry matter are presented based on air-dried weight; others are presented based on dry weight. <sup>b</sup> Means with different letters within the same row are significantly different (p < 0.05). <sup>c</sup> Black ear, *Auricularia mesenteria*; red ear, *A. polytricha*; jin ear, *A. fuscosuccinea* brown strain; snow ear, *A. fuscosuccinea* white strain; silver ear, *Tremella fuciformis*.

**Table 2. Content of Soluble Sugars in Ear Mushrooms** 

		content <sup>b</sup> (mg/g of dry wt)				
sugar	black ear <sup>c</sup>	$\operatorname{red}$ $\operatorname{ear}^c$	jin ear <sup>c</sup>	snow ear <sup>c</sup>	silver ear <sup>c</sup>	
galactose glucose mannose other reducing sugars <sup>a</sup>	ND <sup>d</sup> ND ND 178.10B	1.78B ND 0.79 176.23B	1.75B 0.93 ND 96.04D	1.90A ND ND 107.13C	1.01C ND ND 315.43A	

total 178.10B 178.80B 98.72D 109.03C 316.44A

 $^a$  Other reducing sugars = total reducing sugars – (galactose + glucose + mannose).  $^b$  Means with different letters within the same row are significantly different (p < 0.05).  $^c$  See footnote c of Table 1 for genus/species names of mushrooms.  $^d$  ND, not detected.

snow ear contained the second highest amount (8.51%). The crude protein contents were high in the black and snow ears (15.46 and 12.53%, respectively). The advantage for the snow ear to compete with the silver ear from the standpoint of nutrition values was its lowest carbohydrate and higher fat, fiber, and protein contents. The same advantage could also make it possible to compete with other ear mushrooms.

Auricularia sp. (Philippine var.) contained 4.2% protein, 8.3% fat, 63.0% carbohydrate, 19.8% fiber, and 4.7% ash, based on dry weight (Chang and Miles, 1989). The compositions of Auricularia spp. in Table 1 varied and differed from that of Chang and Miles (1989), in which the species of ear mushrooms analyzed was not identified. The discrepancy among their proximate compositions might be due to the difference in species, strains, the compost used, and growing conditions. The cultivated silver ear contained 7.6% protein, 1.2% fat, 82.7% carbohydrate, 1.3% fiber, and 7.2% ash (Yang, 1988). The composition of the silver ear in this study was similar to that of Yang (1988).

Ear mushrooms contained very low amounts of soluble sugars (Table 2). However, they contained high amounts of other reducing sugars, which were determined according to the dinitrosalicylic acid method. Surprisingly, the silver ear contained the highest amount of other reducing sugars (315.43 mg/g of dry weight). Soluble sugars contained in mushrooms contributed a sweet taste (Litchfield, 1967). However, the results in this study revealed that these ear mushrooms would not give a sweet perception.

The total free amino acid contents in five ears ranged from 0.53 to 1.24 mg/g of dry weight and were in the order of the red, snow, black, silver, and jin ears (Table

**Table 3. Content of Free Amino Acids in Ear** Mushrooms

	content <sup>b</sup> (mg/g of dry wt)				
	black	red	jin	snow	silver
amino acid	$ear^c$	$ear^c$	$ear^c$	$\mathbf{ear}^c$	$\mathbf{ear}^c$
L-alanine	0.15B	0.11B	0.24A	0.04C	0.08BC
L-arginine	0.10BC	0.04C	0.13B	0.08BC	0.46A
L-aspartic acid	0.07B	0.02B	0.12A	0.06B	0.03B
L-glutamic acid	0.27A	0.03C	0.14B	0.16B	0.06BC
glycine	0.02A	0.01A	0.02A	0.02A	0.02A
L-isoleucine <sup>a</sup>	0.01C	0.04A	0.04A	0.03B	0.04A
L-lysine <sup>a</sup>	0.10A	0.13A	0.13A	$\mathrm{ND}^d$	0.11A
L-phenyl- alanine <sup>a</sup>	0.02C	0.04BC	0.07AB	0.02C	0.09A
L-serine	0.05B	0.02B	0.08A	0.16A	0.02B
L-threonine $^a$	0.09AB	0.03B	0.16A	0.16A	0.07B
L-tryptophan <sup>a</sup>	0.01B	0.01B	0.02A	0.01B	0.01B
L-tyrosine	0.02B	0.02B	0.03B	0.06A	0.03B
L-valine <sup>a</sup>	0.02B	0.03B	0.06A	0.03B	0.03B
total	0.93AB	0.53C	1.24A	0.70BC	1.05AB

<sup>&</sup>lt;sup>a</sup> Essential amino acid. <sup>b</sup> Means with different letters within the same row are significantly different (p < 0.05). <sup>c</sup> See footnote c of Table 1 for genus/species names of mushrooms. <sup>d</sup> ND, not detected.

**Table 4. Content of Taste Characteristics of Free Amino** Acids in Ear Mushrooms

	content <sup>a</sup> (mg/g of dry wt)					
taste characteristic <sup>a</sup>	black ear <sup>c</sup>	$\operatorname{red}_{\operatorname{ear}^c}$	$egin{split} \mathbf{jin} \ \mathbf{ear}^c \end{split}$	$\operatorname{snow}$ $\operatorname{ear}^c$	silver ear $^c$	
MSG-like	0.34A	0.05C	0.26B	0.22B	0.09C	
sweet	0.31AB	0.17B	0.50A	0.25B	0.19B	
bitter	0.16C	0.16C	0.32B	0.17C	0.63A	
tasteless	0.12AB	0.15A	0.16A	0.06B	0.14A	
total	0.93AB	0.53C	1.24A	0.70BC	1.05AB	

<sup>&</sup>lt;sup>a</sup> MSG-like, monosodium glutamate-like: Asp + Glu; sweet, Ala + Gly + Ser + Thr; bitter, Arg + Ile + Phe + Try + Val; tasteless, Lys + Tyr. b Means with different letters within the same row are significantly different (p < 0.05). <sup>c</sup> See footnote c of Table 1 for genus/species names of mushrooms.

3). The free amino acids in the highest amounts were glutamic acid for the black ear, lysine for the red ear, alanine for the jin ear, glutamic acid, serine, and threonine for the snow ear, and arginine for the silver ear. Table 4 divides the free amino acids into several classes on the basis of their taste characteristics as described by Komata (1969). Aspartic and glutamic acids were monosodium glutamate-like (MSG-like) components, which gave the most typical mushroom taste, the umami taste, or palatable taste that was the characteristic taste of MSG and 5'-nucleotides (Yamaguchi, 1979). The contents of MSG-like components ranged from 0.05 to 0.34 mg/g and were in the order of the red, silver, snow, jin, and black ears. The contents of sweet components ranged from 0.17 to 0.50 mg/g and were in the order of the red, silver, snow, black, and jin ears. However, the content of bitter components was high in the silver ear.

Chen (1986) conducted a series of sensory evaluations on synthetic mushroom extracts prepared by omitting and adding soluble components and found that alanine, glycine, and threonine (sweet) and aspartic and glutamic acids (MSG-like) were taste-active amino acids in common mushrooms (Agaricus bisporus), whereas none of the bitter components were found to be taste-active. Therefore, MSG-like and sweet components would be responsible for the natural taste of ear mushrooms. Due to the low soluble sugar content, the bitterness from

Table 5. Content of 5'-Nucleotides in Ear Mushrooms

		content <sup>c</sup> (mg/g of dry wt)				
5'-nucleotide <sup>a</sup>	$\begin{array}{c} {\sf black} \\ {\sf ear}^d \end{array}$	$\operatorname{red}_{\operatorname{ear}^d}$	jin ear <sup>d</sup>	$rac{snow}{ear^d}$	silver ear <sup>d</sup>	
5'-AMP	0.22A	0.17A	0.01B	0.17A	$\mathrm{ND}^d$	
5'-CMP	0.42B	0.13B	3.20A	0.30B	0.72B	
5'-GMP	0.05B	ND	0.01C	0.08A	0.01C	
5'-IMP	0.20BC	0.08C	0.78B	1.41A	0.05C	
5'-UMP	0.05B	ND	0.58A	0.51A	0.21B	
5'-XMP	0.15C	0.31C	0.81A	0.68AB	0.93A	
${f flavor}^b$	0.40D	0.39D	1.60B	2.17A	0.99C	
total	1.09D	0.69E	5.39A	3.15B	1.92C	

<sup>a</sup> 5'-AMP, 5'-adenosine monophosphate; 5'-CMP, 5'-cytosine monophosphate; 5'-GMP, 5'-guanosine monophosphate; 5'-IMP, inosine monophosphate; 5'-UMP, 5'-uridine monophosphate; 5'-XMP, 5'-xanthosine monophosphate. <sup>b</sup> Flavor 5'-nucleotides: 5'-IMP + 5'-GMP + 5'-XMP. <sup>c</sup> Means with different letters within the same row are significantly different (p < 0.05). <sup>d</sup> See footnote c of Table 1 for genus/species names of mushrooms. e ND, not detected.

bitter components in ear mushrooms could probably be masked only by sweet components.

The content of total free amino acids in the five ear mushrooms (0.53-1.24 mg/g of dry weight) was much lower than that in common mushrooms (77.92 mg/g) (Tseng and Mau, 1997), in shiitake mushrooms (19.43-35.89 mg/g) (Lin, 1988), in straw mushrooms (Volvariella volvacea) (36.11-60.18 mg/g) (Mau et al., 1997), and in black poplar mushrooms (Agrocybe cylindracea) (39.30-63.34 mg/g) (Mau and Tseng, 1998). The content of MSG-like components in the five ear mushrooms (0.05–0.34 mg/g of dry weight) was much lower than that in common mushrooms (22.67 mg/g) (Tseng and Mau, 1997), in shiitake mushrooms (3.75-9.06 mg/g) (Lin, 1988), in straw mushrooms (11.20-26.21 mg/g) (Mau et al., 1997), and in black poplar mushrooms (10.85–13.05 mg/g) (Mau and Tseng, 1998). Therefore, the taste components of the five ear mushrooms might be much less intense than those mentioned above.

The contents of total 5'-nucleotides ranged from 0.69 to 5.39 mg/g and were found in the order of the red, black, silver, snow, and jin ears (Table 5). Flavor 5'nucleotides were found to be 5'-guanosine monophosphate (5'-GMP), 5'-inosine monophosphate (5'-IMP), and 5'-xanthosine monophosphate (5'-XMP) (Chen, 1986). The contents of flavor 5'-nucleotides were high in the snow and jin ears. 5'-GMP gave the meaty taste (Litchfield, 1967), and the synergistic effect of flavor 5'nucleotides with MSG-like components might greatly increase the umami taste of ear mushrooms (Yamaguchi et al., 1971). The contents of MSG-like components plus flavor 5'-nucleotides were high in the jin and snow ears (Tables 4 and 5), which revealed that the umami taste intensities of the jin and snow ears were similar and stronger than that of the black, red, and silver ears.

Total 5'-nucleotide content in the five ear mushrooms (0.69–5.39 mg/g of dry weight) was slightly higher than that in black poplar mushrooms (0.67-1.51 mg/g) (Mau and Tseng, 1998) and lower than that in shiitake mushrooms (7.26-11.47 mg/g) (Lin, 1988), in common mushrooms (11.35 mg/g) (Tseng and Mau, 1997), and in straw mushrooms (27.01-44.71 mg/g) (Mau et al., 1997). Furthermore, flavor 5'-nucleotide content in the five ear mushrooms (0.39-2.17 mg/g of dry weight) was higher than that in black poplar mushrooms (0.21-0.63 mg/g) (Mau and Tseng, 1998), slightly lower than that in shiitake mushrooms (1.73-3.67 mg/g) (Lin, 1988), and much lower than that in common mushrooms (4.19 mg/g) (Tseng and Mau, 1997) and straw mushrooms (4.42–9.00 mg/g) (Mau et al., 1997). On the basis of the previous results, the five ear mushrooms contained low amounts of MSG-like components and flavor 5′-nucleotides. The synergistic effect of the low amount of flavor 5′-nucleotides with the low amount of MSG-like components might give rise to the less intense umami taste of ear mushrooms different from those mentioned above. However, further sensory evaluation is needed.

Due to their similar appearances, the snow ear, which has a more rubbery texture, currently competes with the silver ear, which has a soft texture. In this study, as compared to the silver ear, the snow ear had higher fat, fiber, and protein contents, lower carbohydrate and reducing sugar contents, higher MSG-like and sweet component contents, and higher flavor 5'-nucleotide contents. Also, the snow ear competes with the black, red, and jin ears due to their similar textures, becaase it is a stable white mutant originated from the jin ear. As compared to the black and red ears, both the jin and snow ears had higher fat and fiber contents, higher MSG-like and sweet component contents, and higher flavor 5'-nucleotide content. From the results shown above, it could be concluded that these five kinds of ear mushrooms, in addition to their characteristic appearances, were considerably different in both their proximate compositions and taste components. However, to determine the relationship of the palatability of the five ear mushrooms with their soluble components, further sensory evaluation is needed.

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