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Nonvolatile taste components of *Grifola frondosa*, *Morchella esculenta* and *Termitomyces albuminosus* mycelia

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Abstract

Three species of mushroom mycelia are commercially available in Taiwan, namely *Grifola frondosa* (maitake), *Morchella esculenta* (morel), and *Termitomyces albuminosus* (termite mushroom), and their nonvolatile taste components were studied. All mycelia were high in contents of carbohydrate, crude fat and protein but low in contents of crude ash and fiber. Arabitol, glucose, mannitol and trehalose were detected in these three mycelia and total amounts of sugars and polyols were 63.69–83.89 mg/g. Total free amino acid contents in three mycelia ranged from 35.67 to 50.37 mg/g. The monosodium glutamate-like components of *G. frondosa* mycelia (6.51 mg/g) was approximately twofold higher than *M. esculenta* and *T. albuminosus* mycelia (3.58 and 3.11 mg/g, respectively). As compared with other components, sweet and bitter components were much higher in these three mycelia. Contents of total 5′-nucleotides in these three mycelia ranged from 13.32 to 26.19 mg/g and were in the descending order of *T. albuminosus M. esculenta* S. *frondosa*. Equivalent umami concentration of *T. albuminosus* mycelia was higher than those of *G. frondosa* and *M. esculenta* mycelia. Overall, three mycelia possessed highly intense umami taste.

Keywords: Grifola frondosa; Morchella esculenta; Termitomyces albuminosus; Soluble sugars; Free amino acids; 5'-nucleotides; Equivalent umami concentration

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1. Introduction

Mushrooms have recently become attractive as functional foods, and a source of physiologically beneficial medicine. Mushrooms are commonly used as food and food-flavoring substance and also a traditional Chinese medicine. Recently, three species, including maitake, morel and termite mushrooms, are highly valued in Taiwan, partially due to their rareness and difficulty in cultivation.

Grifola frondosa (Dickson: Fries) Gray (maitake) is also called the king of mushrooms and the hen of the

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woods (Stamets, 1993). Both fruit bodies and mycelia of maitake have been reported to possess beneficial antitumor and antiviral properties (Mizuno, Ohsawa, Hagiwara, & Kuboyama, 1986). However, this mushroom is not yet available in the fresh market in Taiwan. *Morchella esculenta* (L.: Fries) Persoon (morel) is a mushroom of high gastronomic quality due to its delicate flavor and meaty texture (Phillips, 1991) and has been defying attempts at domestication. Nevertheless, morel is not available in Taiwan.

Termitomyces albuminosus (Berkeley and Broome) Heim (termite mushroom), also called chicken julienne mushroom, is a symbiotic fungus found in tropical Africa and Asia (Abe & Matsumoto, 1979; Wood & Sands, 1978). Termites cultivate this fungus in their nests as food (Heim, 1977). The fruit bodies form inside

the tunnels and bore through the very hard layer of inert matter, forcing their way through it with a special umbo (Kendrick, 2001). This rare termite mushrooms is also found in Taiwan and is extremely tasty. Like morel, termite mushrooms are not always available.

In addition to dried mushrooms imported from China or Japan, alternative or substitute mushroom products are mycelia of these three mushrooms, mainly prepared from submerged culture. These mycelia are used as food and food-flavoring materials, and also in the formulation of nutraceutical and functional foods. The antioxidant properties and antioxidant components of these three mycelia have been studied (Mau, Chang, Huang, & Chen, 2004). As a food ingredient, the chemical composition and taste components of these mycelia may correlate with their product acceptability. However, the information is not available. Therefore, our objective was to examine the nonvolatile taste components in these mycelia of three mushrooms, including their proximate compositions, soluble sugars, free amino acids and 5'-nucleotides. Equivalent umami concentrations (EUC) of these mycelia were also evaluated.

2. Materials and methods

2.1. Mushroom mycelia

Freeze-dried mycelia of *G. frondosa*, *M. esculenta* and *T. albuminosus* were obtained from the Biotechnology Center, Grape King Inc., Chungli City, Taiwan. For each mycelium, three dried samples (~50 g each) were randomly selected and ground using a mill (Retsch ultracentrifugal mill and sieving machine, Haan, Germany) to obtain fine powder (60 mesh).

2.2. Proximate analysis

The proximate compositions of these three species of mushroom mycelia, including moisture, crude ash, crude fat, crude fiber and crude protein, were determined according to the methods of AOAC (1990). The nitrogen factor used for crude protein calculation was 4.38 (Crisan & Sands, 1978). The carbohydrate content (mg/g) was calculated by subtracting the contents of crude ash, fat, fiber and protein from 1000 mg/g of dry matter and expressed as mg/g of dry weight. Total reducing sugars were determination using the 3,5-dinitrosalicylic acid (DNS) method as described by James (1995). The absorbance of each sample solution was measured at 540 nm on a Hitachi 2001 spectrophotometer. Total reducing sugars were calculated based on a calibration curve of glucose.

2.3. Soluble sugar and polyol assay

Soluble sugars and polyols were extracted and analysed as described by Ailouni, Beelman, Thompson, and Mau (1995). Freeze-dried mushroom mycelia (600 mg) were extracted with 50 ml of 800 ml/l aqueous ethanol (95% pure, Taiwan Tobacco & Wine Monopoly Bureau, Taipei). 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 800 ml/l 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 Millex-HV filter unit (13 mm, Millipore, Billerica, MA, USA), and filtered using a 0.45-µm PVDF filter (Millipore) prior to injection onto high-performance liquid chromatograph (HPLC).

The HPLC system consisted of, a Shimadzu LC-10AT VP pump, a Rheodyne 7725i injector, a 20-µl sample loop, a Shimadzu RID-10A detector, and a Phase Sep-NH $_2$ column (4.6 × 250 mm, 5 µm, Phase Separation Inc., Norwalk, CT, USA). The mobile phase was 85 ml acetonitrile (LC grade, Tedia Co., Fairfield, OH, USA)/15 ml deionized water at a flow rate of 1.0 ml/min. Each sugar or polyol was identified using the authentic sugar or polyol (Sigma Chemical Co., St. Louis, MO, USA) and quantified by the calibration curve of the authentic compound.

2.4. Free amino acid assay

Freeze-dried mushroom mycelia (500 mg) was shaken with 50 ml of 100 mmol/l 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 Millex-HV filter unit (13 mm), and filtered using a 0.45-μm PVDF filter. This filtrate was mixed with *o*-phthalaldehyde reagent (Sigma Chemical Co., St. Louis, MO, USA) in an eppendorf tube, shaken to facilitate derivatization and then immediately injected onto HPLC.

The HPLC system was the same as for sugar and polyol analysis but included a Hitachi L-7485 fluorescence detector with fluorescence excitation at 340 nm and emission at 450 nm, and a LiChrospher 100 RP-18 column ($4.6\times250\,\mathrm{mm}$, 5 µm, Merck, Darmstadt, Germany). The mobile phases were A, 50 mmol/l sodium acetate (pH 5.7) containing 50 ml/l tetrahydofuran; B, deionized water; and C, methanol. The gradient was A:B:C 80:0:20 (v/v/v) to 33:0:67 for 0–38 min, 0:33:67 for 38–40 min, and 0:100:0 for 40–43 min and the flow rate was 1.2 ml/min (Mau, Chyau, Li, & Tseng, 1997). Each amino acid was identified using the authentic amino acid (Sigma) and quantified by the calibration curve of the authentic compound.

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