# **Detection of Taurine in Basidiomycetes**

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Abstract – Taurine is one of the essential amino acids for humans and many of mammals. It is produced and contained in fishes, shells, plants and algae, but has never been found in fungi. We examined six mushrooms for taurine and detected taurine in five of them. Taurine was determined by an automated amino acid analyzer using ion-exchange chromatography, being eluted between phosphoserine as a distinct peak on the chromatogram. Fruit bodies of *Flammulina velutipes* contained 83 µmoles/100 g fresh wt, the highest level among them, *Agaricus bisporos* 65 µmoles, *Lentinus edodes* 49 µmoles, *Pleurotus ostreatus* 9 µmoles, and *Auricularia auricula-judae* 20 µmoles. Taurine was not detected in *Ganoderma lucidum*. As far as fungi are concerned, this is the first report of the detection of taurine in Basidiomyces.

Key words 
Taurine content, basidiomycetes, amino acid analyzer, mushrooms

Taurine (2-aminoethanesulfonic acid) is a sulfur-containing amino acid which was first identified in the bile of an ox by Tiedemann and Gmelin (1827). As with other amino acids, taurine has a small molecular weight of 125.1 and exists as a zwitterion in the physiological condition. The roles of taurine have been demonstrated throughout the animal body, notably in the heart, brain, reproductive system, muscle and eye (Huxtable, 1992). The most prominent functions of taurine are antioxidation (Trachtman et al., 1993), hepatoprotection (Timbrell et al., 1995), intracellular calcium modulation (Militante and Lombardini, 1999), neuromodulation (Ferco and Bobyock, 1988), membrane stabilization and osmoregulatory action (Huxtable, 1992). Yet, it has certain chemical and biological properties that make it unique among the amino compounds: 1) taurine is a β-amino acid, which designates that the amino group is attached to the β-carbon of the structure; 2) a sulfonic acid group provides the acidic group rather than a carboxylic group which is commonly present in amino acids. Since the sulfonic acid group is more acidic (pKa=1.5) than a carboxyl group (pKa=2.1), taurine remains ionizd at a lower pH than amino acids bearing a carboxyl group; 3) taurine is not incorporated into any protein. Even though it forms a few peptides such as glutarine (gamma-L-glutarnyl-taurine) in brain and parathyroid tissues (Feuer et al., 1978; Csaba et al.,

1979), most taurine in animal tissues and biological fluids remains as a free amino acid.

It is produced and contained in fishes, shells, plants and algae, but has never been found in fungi. We examined six mushrooms for taurine and detected taurine in five of them. Carpophores of *Flammulina velutipes* contained 83 µmoles/ 100 g fresh wt, the highest level among them, *Agaricus bisporus* 65 µmoles, *Lentinus edodes* 49 µmoles, *Pleurotus ostreatus* 9 µmoles, and *Auricularia auricula-judae* 20 µmoles. However, taurine was not detected in *Ganoderma lucidum*. As far as fungi are concerned, this is the first report of the detection of taurine in Basidiomyces.

## MATERIALS AND METHODS

## Preparation of sample

For the analysis of taurine concentration in basidiomycetes, six commonly used mushroom samples (*Flammulina velutipes*, *Agaricus bisporus*, *Lentinus edodes*, *Auricularia auricula-judae*, *Pleurotus ostreatus*, and *Ganoderma lucidum*) were obtained commercially from three different grocery stores located in Seoul. A portion of sample was homogenized in ice-cold 0.05 M potassium phosphate buffer (pH 6.8) using a polytron homogenizer (M133/128-O, Biospec Products Inc., Bartlesville, OK, USA) to form a 2.5~20% (w/v) homogenate. The homogenates were centrifuged at 20,000 ×

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g for 30 minutes at  $4^{\circ}$ C, and the supernatant was deproteinized using 20% sulfosalicylic acid, and filtered through 0.2  $\mu$ m filter (PVDF Acrodisc 13, Gelman Sciences).

#### Analysis by amino acid analyzer

Taurine concentration in the filtrate was determined by an automated amino acid analyzer (Biochrom 20, Pharmacia LKB Biotech, Cambridge, England) based on ion-exchange chromatography (Moore and Stein, 1963). The 20 µl aliquot of the filtered sample was injected into the lithium high performance column (90 × 4.6 mm, Pharmacia LKB Biotech). The taurine peak was obtained at about 7.0~7.3 min with 0.2 M lithium citrate buffer, pH 2.8 and 0.3 M lithium citrate buffer, pH 3.0 as mobile phases running at a flow rate of 25.0

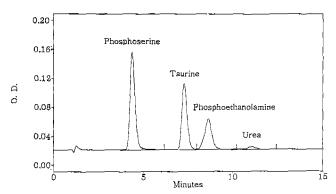
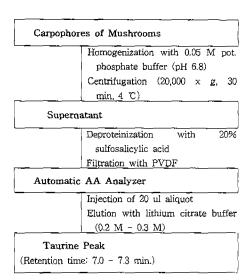


Fig. 1. Chromatogram of standard taurine with automated amino acid analyzer. The retention time of standard taurine was 7.3 min in lithium high performance column ( $90 \times 4.6$  mm, Pharmacia LKB Biotech). The eluated taurine was reacted with ninhydrin and detected at 570 nm.



**Scheme I.** Analysis of taurine in the carpophores of Basidiomycetes with automated amino acid analyzer.

ml/hour for 2 min and 12 min, consecutively at 34 °C (Fig. 1). The eluted amino acids were reacted with ninhydrin and detected at 570 nm (Scheme I).

Standard solution of amino acids (#A6407, Sigma Co., St. Louis, MO, USA) was used to calibrate the instrument to insure accuracy in the qualitative and quantitative measurements of amino acids present in our samples. Taurine recoveries from taurine standard solutions (8~180 nmol/mL) and the sample with a known amount of taurine added were found to be from 97.5 to 102%. Taurine concentration was expressed as a mean  $\pm$  standard error of triplicate samples for each species of the mushrooms.

### RESULTS AND DISCUSSION

By the automated amino acid analyzer using ion-exchange chromatography, taurine was eluted between the two strong acidic amino acids, phosphoserine and phosphoethanolamine as a distinct and symmetrical peak on the chromatogram. Most of the mushrooms tested in the present study contained taurine in the range of 3.7~83  $\mu$ moles/100 g fresh wt. Carpophores of Flammulina velutipes contained the highest level of taurine (83.2  $\pm$  6.6  $\mu$ moles/100 g fresh wt) among the commonly used mushrooms (Fig. 2). Lentinus edodes and Agaricus bisporus also contained high levels of taurine, in the concentrations of 48.6  $\pm$  5.4 and 64.9  $\pm$  3.5  $\mu$ moles/100 g fresh wt, respectively. Pleurotus ostreatus contained 8.9  $\pm$  0.65  $\mu$ moles taurine/100 g fresh wt. Dried samples of Auricu-

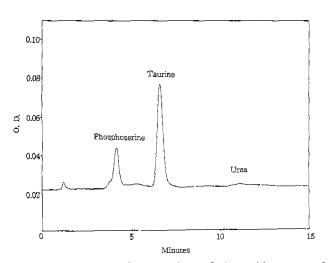


Fig. 2. Chromatogram of Flammulina velutipes with automated amino acid analyzer. Fresh carpophores of the mushroom was used for the preparation of sample. Taurine was detected at the retention time of 7.0 min in lithium high performance column  $(90 \times 4.6 \text{ mm}, \text{Pharmacia LKB Biotech})$ .

Table I. Taurine concentrations in commonly used mushrooms<sup>c</sup>

Basidiomycetes	Korean name	μmoles/100 g wt	mg/100 g wt
Flammulina velutipes <sup>a</sup>	Paengibeoseot	$83.2 \pm 6.6$	$10.4 \pm 0.8$
Agaricus bisporus <sup>a</sup>	Yangsongi	$64.9 \pm 3.5$	$8.1 \pm 0.4$
Lentinus edodes <sup>a</sup>	Pyogobeoseot	$48.6 \pm 5.4$	$6.1 \pm 0.7$
Pleurotus ostreatus <sup>a</sup>	Neutaribeoseot	$8.9\pm0.6$	$1.1 \pm 0.1$
Auricularia auricula-judae <sup>b</sup>	Mokibeoseot	$19.5 \pm 2.1$	$2.4 \pm 0.3$
Ganoderma lucidum <sup>b</sup>	Yongjibeoseot	ND	ND

<sup>&</sup>lt;sup>a</sup>Fresh carpophores

Values are mean ± standard error of triplicate samples

ND: not detected

laria auricula-judae contained  $19.5\pm2.1$  µmoles taurine/100 g wt, which are estimated as 3.3 µmoles taurine per 100 g of the fresh sample based on the assumption that water contents of dried and fresh mushrooms are approximately 15% (National Rural Living Science Institute, 1996). On the contrary, taurine was not detected at all in *Ganoderma lucidum* in this study.

Lägdesmäki (1986) noted that the fraction of taurine and phosphoethanolamine of the plant tissue extracts overlapped seriously from the ion-exchange chromatography system. This problem appears to be solved completely in our chromatography system. Kataoka and Ohnishi (1986) measured the taurine content in plant tissues by gas chromatography with electron-capture detection (GC-ECD), and demonstrated that seaweeds contained high levels of taurine, in the range of 10~1000 nmol/g wet weight, while taurine contents of land plants were much lower than this. There are millimolar concentrations of taurine in mammalian tissues, but only micromolar or less in many higher plants, molds and bacteria, although taurine is widely distributed in these organisms (Jacobson and Smith, 1968; Lägdesmäki, 1986; Kataoka and Ohnishi, 1986; Pasantes-Morales et al. 1989). Our recent findings (Park et al., 1998) also confirmed that taurine is frequently detected in plant kingdom in much lower concentrations  $(10^{-2} \sim 10^{-3}$ -folds) than those found in marine organisms and mammals. Taurine content in edible mushrooms appears to be relatively high as compared to that of other plants, although it varies greatly depending on the species of mushroom. Flammulina velutipes showed the highest taurine content among the land plants tested so far. Recent studies in rats (Park et al., 1997; Gandhi et al., 1992) demonstrated that taurine supplementation significantly lowered plasma and hepatic cholesterol and triglyceride concentrations. Mushrooms are excellent candidates for a high taurine/low cholesterol food, and this could partly explain the hypolipidemic action of certain mushrooms, eritadenin (Sugiyama et al., 1995) and Lentinus edodes (Sugiyama et al., 1993), which has been reported to be mediated by a modification of hepatic phospholipid metabolism. This is the first report of the detection of taurine in Basidiomycetes.

#### **ACKNOWLEDGEMENTS**

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<sup>&</sup>lt;sup>b</sup>Dried carpophores

<sup>&</sup>lt;sup>c</sup>Concentration of taurine was determined by automated amino acid analyzer using ion-exchanger chromatography.

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