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DETERMINATION OF VOLATILE AROMA COMPOUNDS OF *GANODERMA LUCIDUM* BY GAS CHROMATOGRAPHY MASS SPECTROMETRY (HS-GC/MS)

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Abstract

This study was conducted at Horticulture Department of Cukurova University, Adana, Turkey during 2010-2011. Fresh sample of *Ganoderma lucidum* collected from Mersin province of Turkey was used as material. Volatile aroma compounds were performed by Headspace Gas Chromatography (HS-GC/MS). Alcohols, aldehydes, acids, phenol, L-Alanine, d-Alanine, 3-Methyl, 2-Butanamine, 2-Propanamine were determined. 1-Octen-3-ol (Alcohol) and 3-methyl butanal (Aldehyde) were identified as major aroma compounds.

Key words: Ganoderma, volatiles, headspace, gas chromatography-mass spectrometry.

Introduction

Ganoderma lucidum (Curtis) P. Karst. called "mushroom of immortality" and "Reishi" in Turkey is one of the significant and valuable medicinal mushrooms and it is consumed as tea or medicine (Yakupoğlu and Pekşen, 2011). This mushroom is known with different names in different countries in the world such as "Ling Chi, Ling Chih or Ling Zhi' in China and Korean, "Reishi or Mannentake" in Japan (Stamets, 2000) and "Ganoderma" in USA. Medical usage of *Ganoderma* was indicated by many researchers. It was known to use diseases such as heart diseases, chronic bronchitis, asthma, hypertension, hepatitis, hypercholesterol, allergy and cancer (Hobbs, 1944; Chen and Zhang, 1987). Triterpenes and polysaccharides are two of important chemical compounds in *Ganoderma*. Polysaccharides, especially beta glucans, stimulate immune system and beta glukans induce production of T cells that against infected cells (Stamets, 2000). Ganoderic acid prevents to coagulation in blood, reduce cholesterol level in blood (Stamets, 2000; Morigawa et al., 1986) and regulate blood pressure, lipid (Stamets, 2000; Kabir et al., 1988) and glucose level (Stamets, 2000; Kimura et al., 1988) of blood. Also it is known as a diuretic, laxative, sedative and tonic (Hobbs, 1944; Liu and Bau, 1980). Volatile flavor compounds from submerged cultured *Ganoderma sinense* (J.D. Zhao, L.W. Hsu & X.Q. Zhang**)** mycelium were analysed by Headspace Gas Chromatography (HS-GC/MS) by Wang et al., (2009). Different types of compunds; including ketones, esters, lactones, alcohols, aethers and hyrodroxybenzenes were identified in *G. sinense*. However, to best our knowledge there is no previous studies on volatile compounds of fresh *G. lucidum* samples. For this purpose, we aimed to detect volatile compounds of *G. lucidum* using HS-GC/MS technique.

Material and Methods

G. lucidum specimens collected from Mediterrenean Region (Mersin province) of Turkey were used in this study. After surface sterilization of this mushroom specimens in sterile bench, small pieces from internal region of fruit body were cut and placed into nutrient medium. Potato Dextrose Agar (PDA: Potato extract: 4 g/l, Dextrose: 20 g/l, Agar: 15 g/l and pH:7.5) was used as nutrient medium. Sterilization of nutrient medium were performed in 121°C in 1.2 atm for 15 min using autoclave. Mycelial growth in all of petri dishes were obtained in 8 days. Then mycelia in each petri dish were divided 6-8 pieces and inoculated to sawdust plastic culture bags (made of high-pressure-resistant plastic) for fruiting body grown. To prepare sawdust culture: firstly oak sawdust material was wet with water to provide 70% humidity, then added wheat bran, zeolite and mixed. This mixture was filled plastic bags and then sterilized in 121°C in 1.2 atm for 45 min using autoclave. All of sawdust cultures bags were placed in mushroom growing room has 25°C temperature, 80% humidity and 1000 lux light. Harvested *G. lucidum* samples were used as fresh for aroma analyses adding 5 g NaCl for 50 g of sample.

Volatile aroma analyses were done at Horticulture Department of Çukurova University, Adana-Turkey. Volatile compounds were analyzed on an HS/GC/MS apparatus equipped with an HP-5 MS (30 m × 0.25 mm × 0.25 µm) fused-silica capillary column. Helium (1 ml/min) was used as a carrier gas. The SPME holder, for manual sampling, and fibres used in this study were purchased from Supelco (Bellefonte, PA). The polydimethylsiloxane (PDMS) 100 mm fibres were used and the fibres conditioned for 1 h at 240°C in the GC injector port before being used, and they were cleaned between analyses to prevent contamination. The injector temperature was 250°C, set for split less injection. The oven conditions were set to 50°C for 1 min and then the temperature was increased to 200°C at a rate of 4°C/min. Thermal desorption was allowed for 1.5 min. The detector

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temperature was 280°C. The components were identified by comparison of mass spectra and retention time data complemented with a Wiley, Flavor and NIST GC-MS libraries.

Results and Discussion

18 aroma compunds were identified in this study and obtained results were given in Table 1. As seen in Table 1, alcohols such as 1-Octen-3-ol, 3-Octanol, 1-Octanol, 2-ethyl -1-Hexanol were detected as the major compounds and consisting of approximately 48.05% of total aroma. Among the detected alcohols 1-Octen-3-ol was detected as the major one. 1-Octen-3-ol (mushroom, butter, resinous), 3-Octanol (fruity, cod liver oil, citrus, weakly nutty, fungal), 1-Octanol (fruity-flowery, sweet soap, orange, waxy, sweet) 1-Octen-3-one (boiled mushrooms, metallic, fungal, wild mushroom), 3-Octanone (fruity, sweet, musty, floral, lavender, sweet ester), 2-Octen-3-ol and 3-Octanal are the well-known and common aroma compounds (Jong and Birmingham, 1993; Taylor and Linforth, 2010). The identified aldehydes were detected as 3-methyl butanal, 2-Octenal, pentanal, 2,4-Decadienal, 1-Proponal. Among them 3-methyl butanal was detected as the major aldehyde. As for the acids; 1,2-Benzenedicarboxylic acid, Hexadecanoic acid, Octadecanoic acid were detected and among them hexadecanoic acid was the major. Hexadecanoic acid is a synonym of palmitic acid and it used to produce soap and cosmetic. 3-Octanone, phenol and other compounds such as d-Alanine, L-Alanine, 3-Methyl, 2-Butanamine, 2-Propanamine were detected also. Phenolic compounds are found as major antioxidant componets in mushrooms (Tsai et al., 2009; Lee et al., 2010).

In addition to Benzeneacetaldehyde, Cyclooctene, Formic acid, Benzothiazole 2,4-Decadienal, 2,6-di-butyl-2,5-cyclohexadiene-1, Phythalic acid, Ethanol, 2-Heptanamine, Alanine, Cyclopentane, Butanal, Eicosane, Squalene, Iron, Tetradecane, Nonane, Decanal, 2H-Pyran-2-One, Oxirane, n-Hexylmethylamine, 2-Methylaminoethanol, 1,2-Ethanediamine, Cyclohexadecane, Heptadecene were detected trace amount in *G. lucidum*.

Table 1: Volatile composition of *Ganoderma lucidium* by HS-GC/MS.

R.T	Compound name	Area %
Alcohols		
7.35	1-Octen-3-ol	34.67
7.50	3-Octanone	14.41
7.72	3-Octanol	9.72
8.61	2-ethyl 1-Hexanol	3.08
9.76	1-Octanol	0.58
Aldehydes		
6.40	3-methyl Butanal	18.87
9.41	2-Octenal	0.85
1.45	1-Proponal	0.28
2.48	Pentanal	0.23
16.22	2,4-Decadienal	0.07
Acids		
32.28	Hexadecanoic acid	2.38
35.42	Octadecanoic acid	0.96
30.47	1,2-Benzenedicarboxylic acid	0.16
Phenol		
22.20	Phenol	4.34
Other compounds		
1.89	L-Alanine	0.21
1.90	d-Alanine	0.10
3.20	3-Methyl, 2-Butanamine	0.04
3.26	2-Propanamine	0.02
	Other compounds	9.03

RT: Retention time

28 volatile flavor compounds were identified in *G. sinense* by Liu et al., (2009). Also 26 volatile compounds were detected in *G. sinense* by Wang et al., (2009). In both of these studies, ketones, alcohols and lactones were found as main compounds. In a study conducted by Chen et al., (2010), 58 compunds were determined in *G. lucidum* mycelia and 1-octen-3-ol, ethanol, hexanal, 1-hexanol, sesquirosefuran, 3-octanol, 3-octanone were found as main volatile flavor compounds. In all of these studies, mycelia were used to determine volatile aroma compounds. Presented in this study, the fresh mushroom samples collected from nature and cultured were used for aroma analyses. Alcohols and aldehydes were identified as main volatile aroma compounds.

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