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Research Article

LIPID COMPOSITIONS OF *Bjerkandera adusta* (WILLD) P KARST

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ABSTRACT

Mushrooms are considered as a substantial nutrient, which are the source of many vitamins as well as protein and mineral. Since ancient times, it has been known that mushrooms used as food as well as medicine in many societies. In addition to the nutritional properties of mushrooms, it is especially preferred by people due to its flavor and aroma. Despite the low fat content of mushrooms, the high proportion of polyunsaturated fatty acids further increasing the importance of mushrooms for nutrition. In this study, fatty acid components of an edible mushroom *Bjerkandera adusta* was investigated. It was determined that 20 of the 26 componets were fatty acid in the lipid fraction of *B.adusta* by using GC and GC/MS systems. Linoleic acid (37.25%), oleic acid (9.51%) and stearic acid (4.51%) were found as major fatty acids of *B.adusta*. Additionally, ergosterol (12.46%) was found in high concentration in the lipid fraction of *B. adusta*. As a result, it was found that the percentage of unsaturated fatty acids in the total lipid fraction is 52.69% in *B.adusta*.
Keywords: *Bjerkandera adusta*, fatty acid, ergosterol, GC, GC/MS, mushroom.

1. INTRODUCTION

In the past, mushrooms have known as a "plant", but later on it has recognized seperate kingdom due to its different characters from plants and animals. Although, many mushroom species are poisonous, some of them have medicinal usage. Some of the mushroom species particularly known as "parasol mushrooms" are used as food. [1-3]. Mushrooms are widely consumed as a food in many countries and it is one of the essential foods due to its impressive tastes, aromas and nutritional properties. Mushrooms are valuable food items due to having high protein, fiber, vitamins, minerals and low fat content [4-6].

Mushrooms have attracted the attention of researchers in recent years, according to their biological activities. Such as; The immunomodulatory, antitumor as well as antiviral, antimicrobial, antimutagenic, antihypertensive, antiinflammatory, antiallergic activities are besides the biological activities of mushrooms [7-8].

Up to date mainly, Lecthin, polysaccharides, polysaccharide-peptides, polysaccharide-protein types of compounds have been isolated from the mushrooms which are these substances which are exhibiting immunomodulatory, anticancer and antioxidant activities [7-9].

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In addition to medicinal properties of mushrooms, there is an increasing of importance on the utilization of them as a "functional food" due to their unique composition including proteins, amino acids, vitamins, minerals and high-content polyunsaturated fatty acid. Although the fat content of mushrooms account about 5%, and 75% of it consists of unsaturated fatty acid components, that makes mushrooms significant part of healthy nutrition. It is known that, especially polyunsaturated fatty acids, omega-3, and -6 series are necessary for human health in the prevention and treatment of hypertension, coronary artery disease, diabetes, osteoporosis, arthritis, cancer, other inflammatory and autoimmune disorders. Due to the medical properties of polyunsaturated fatty acids and their role in natural nutrition, scientific studies on mushrooms are increasing day by day [10-17].

In the literature, there is a short report on fatty acid composition of some mushroom species which are growing on permafrost conditions. According to that paper, linoleic acid, eicosanoic acid, oleic acid and stearic acid have been found major components of *B.adusta* respectively [18].

The aim of this work was to investigate the lipid composition of *Bjerkandera adusta* which is edible and growing naturally in Turkey using by GC-FID and GC-MS systems.

2. MATERIALS AND METHODS

2.1. Mushroom Material

B.adusta is growing naturally in Turkey and known as tree mushrooms due to, growing on tree barks. *B.adusta* was collected from sweetgum (*Liquidambar orientalis*) forests in Köyceğiz-Muğla, province of Turkey in 2015. They were identified and deposited in the Fungarium of Department of Biology, Muğla Sıtkı Koçman University and were stored at -18 ºC until analyses (Fungarium no: AT-2447).

2.2. Chemicals and Instruments

Boron trifluoride-methanol solution (BF3:MeOH), n-hexane and chlroform were obtained from E. Merck (Darmstadt, Germany). GC-FID analyses were performed on a Shimadzu GC-17 AAF, V3, 230 V series gas chromatography (Japan) and GC–MS analyses were on Varian Saturn 2100 (USA).

2.3. Extraction

Dry mushroom sample (50 g) was extracted with 100 mL n-hexane:chloroform (90:10) solvent system for three times (24 h x 3) at room temperature, then filtered and evaporated to dryness under vacuum. They stored at refrigerator (+4^{o}C) until methylation process.

2.4. Methylation of lipid extract of *B.adusta*

The lipid extract (100 mg) was dissolved in 0.5 M NaOH (2 mL). After that solution was heated in a water bath (50 °C), then 2 mL BF3:MeOH was added. The mixture was boiled for 3 minutes, and then the mixture was left until it cooled down, and then the volume was completed to 25 mL with saturated NaCl solution. Esters were extracted with n-hexane; thus, the organic layer was separated. The hexane layer was dried with anhydrous Na_2SO_4 and filtered. The organic solvent was removed under reduced pressure by a rotary evaporator to give methyl esters.

2.5. Gas chromatog raphy (GC) Analysis

GC analysis of the methyl derivatives of lipid fraction of *B.adusta* was performed by Shimadzu GC-17 AAF, V3, 230 V series gas chromatography (Japan) coupled with a Flame Ionisation Detector (FID) and a DB-1 fused silica capillary non-polar column (30 m x 0.25 id., film thickness 0.25 µm). Injector and detector temperatures were 250 and 270 °C, respectively, carrier gas was He at a flow rate of 1.4 mL/min; sample volume, 0.4 µL; split ratio, 50:1. The initial oven temperature was held at 100 °C for 5 min, then increased up to 238 ºC with 3 ºC/min increments and held at this temperature for 9 minutes. The relative percentages of separated compounds were calculated by using GC Solution computer program.

2.6. Gas chromatography-Mass spectrometry (GC-MS) Analysis

GC-MS analysis of the methyl derivatives of lipid fraction determined using by Varian Saturn 2100T (USA) coupled with an ion trap mass spectrometer and a DB-1 MS fused silica non-polar capillary column (30 m x 0.25 mm ID, film thickness 0.25 µm). For GC–MSD detection, an electron ionization system with ionization energy of 70 eV was used. Carrier gas was helium (15 psi) at a flow rate of 1.3 mL/min. Injector and MS transfer line temperatures were set at 250 and 200 °C, respectively. The oven temperature was held at 100 °C for 5 min, then increased up to 238 °C with 3 °C/min increments and held at this temperature for 9 minutes. 0.2 µL was injected manually in the split mode. Split ratio was 50:1. EI-MS were taken at 70 eV ionization energy. Mass range was from m/z 28 to 650 amu. The library search was carried out using NIST and Wiley 2005 (Gas Chromatography-Mass Spectrometry) GC-MS libraries. FAME (Fatty acid Methyl Ester) mixture (Supelco™ 37, Catalog no: 47885-U) were identified by comparing their retention times with those of the pure FAMEs standards.

3. RESULTS AND DISCUSSIONS

The yield of lipid fraction was found 2.50% in *B. Adusta*. The presence of twenty six components were determined in the lipid compositions of *B.adusta* (Table 1). Linoleic acid (C18:2 ω6) (37.25%), ergosterol (12.46%) and oleic acid (C18:1 ω9) (9.51%) were the major lipit components of *B.adusta*. The lipid components in the mushroom were observed to be rich in fatty acids and unsaturated fatty acid amounts were higher than saturated ones in *B.adusta*. Linoleic acid and oleic acid are very important for human diet. These compounds are known to be decreasing the risk of cardiovascular disease and also recommended for the balancing high blood cholesterol.

Besides fatty acids, Ergosterol (12.46%) was one of the major components. Ergosterol is known as provitamin D_2 and mostly found in mushrooms because it is a part of their cytoplasmic membrane. Ergosterol is converted to vitamin D_2 by exposure to ultraviolet (UV) light. Vitamin D_2 is a type of vitamin D and used as prevent and treat vitamin D deficiency. Vitamin D is important for dietary supplement as a regulator of the metabolism of calcium and phosphate [19]. There is some reports on medicinal activities of ergosterol such as; growth-inhibition of bladder cancer, lipid peroxidation, antihyperlipidemic and anti-inflammatory activities [20-23] .

We found also unusual components for mushrooms such as, cinnamic acid (5.13%) and cinnamyl alcohol (0.65%). On the other hand, cinnamic asid derivatives are major constituents of resin of *Liquidambar orientalis* [24]. The *L. orientalis* (oriental sweetgum) is a tertiary period relict endemic taxon of the east Mediterranean [25]. Due to, *B.adusta* is growing on *Liquidambar orientalis,* cinnamic acid and cinnamyl alcohol might be derived from *Liquidambar orientalis*.

Table 1. Lipid Composition (%) of *Bjerkandera adusta*

Peak no	Compound	Concentration (%)
1	Cinnamyl alcohol	0.65
2	Mandelic acid	0.57
3	Cinnamic acid	5.13
4	9-oxo-nonanoic acid ($C_{9:0}$)	0.63
5	10-methyl-undecanoic acid ($C_{11:0}$)	0.34
6	2-oxo-cyclohexane propanoic acid	1.32
7	12-Methyl, tridecanoic acid ($C_{13:0}$)	1.51
8	10-Pentadecen-1-ol	0.26
9	Pentadecanoic acid ($C_{15:0}$)	3.60
10	Palmitic acid ($C_{16:0}$)	3.14
11	Methyl, 3- (3,5-ditertbuthyl-4-hydroxy-phenyl) propionate	1.23
12	3,15-Octadecadien-1-ol acetate	1.18
13	Margaric acid ($C_{17:0}$)	0.83
14	**Linoleic acid ($C_{18:2}$)**	**37.25**
15	**Oleic acid ($C_{18:1}$)**	**9.51**
16	Stearic acid ($C_{18:0}$)	4.51
17	Eicosanoic acid ($C_{20:0}$)	0.88
19	Heneicosanoic acid ($C_{21:0}$)	0.37
18	Docosatetraenoic acid ($C_{22:4}$)	2.79
20	Behenic acid ($C_{22:0}$)	1.65
21	Diisooctyl phthalate	1.35
22	Tricosanoic acid ($C_{23:0}$)	1.31
23	15-tetracosanoic acid ($C_{24:1}$)	3.14
24	Tetracosanoic acid ($C_{24:0}$)	2.89
25	**Ergosterol**	**12.46**
26	Pentacosanoic acid ($C_{25:0}$)	1.50
	Saturated fatty acids	**23.16**
	Unsaturated fatty acids	**52,69**
	Other components	**24.15**

4. CONCLUSIONS

Mushrooms are the valuable and healthy foods in human diet, due to their low-fat composition, low calories and high essential fatty acid levels. The present study indicates that *B.adusta* contains high percentage of unsaturated fatty acids especially linoleic acid and oleic acid. Mushrooms are known to be a good source of vitamin D_2. It contains high percentage of ergosterol (provitamin D2) that has medicinal properties and also phenolic organic compounds were found such as cinnamic acid and cinnamyl alcohol in the lipid fraction of *B.adusta* as well. These results showed that, *B.adusta* has a great potential to use as a supplementary food or nutraceuticals.

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