

Phytochemical and Mineral Analysis of 12 Cultivated Oyster Mushrooms (Pleurotus Species)

Mary Obodai¹ Ebenezer Owusu² Gladys O. Schiwenger² Isaac K. Asante^{2*} Matilda Dzomeku¹

1.CSIR-Food Research Institute, P.O.Box M20, Accra, Ghana

2.Department of Botany, University of Ghana, Legon, Ghana

*Email of corresponding author: asanteisaack@yahoo.com

The research is financed Office of Research and Innovative Division of the University of Ghana, Legon

Abstract

Phytochemical analysis was conducted on methanolic extracts of 4 oyster mushrooms species (*Pleurotus pulmonarius*, *P. ostreatus*, *P. sapidus* and *P. citrinopileatus*) made up of 12 different strains. The study was carried out to determine the contents of total phenol and flavonoids, beta carotene and lycopene, antioxidant properties and concentrations of mineral elements and heavy metals in the macrofungi methanolic extracts. Total phenolic contents ranged from 1.48 µg GAE/g to 3.58 µg of GAE/g with a mean of 2.5 µg of GAE/g and 0.36 coefficient of variation. The 12 strains were statistically different from each other. Total flavonoid content varied from 0.20 µg of RE/g to 2.03 µg of RE/g of dry weight of extracts with a grand mean of 0.85 µg of RE/g and 9.89 coefficient of variation. There was significant difference among the 12 mushroom strains ($P < 0.001$). Beta carotene content varied from 0.60 to 11.46 µg/g and that of lycopene varied from 0.04 to 4.28 µg/g. The IC_{50} values ranged between 43.21 µg/ml and 52.03 µg/ml, the inhibition of DPPH radical by *P. sapidus* strain PSC-1 and *P. ostreatus* strain POA-5 exhibited the highest activity. Significant difference was observed among the strains. Mg content varied from 660.0 mg/kg to 1993.7 mg/kg, Fe content ranged from 349.0 mg/kg to 1374.0 mg/kg and could not be detected in *P. ostreatus* strain POA-7. Ca content ranged from 22.0 mg/kg to 415.3 mg/kg, Mn content varied from 10.7 mg/kg to 48.3 mg/kg and was below detection in the six strains of *P. pulmonarius* strain PPA-2, *P. ostreatus* strain POA-5, *P. ostreatus* strain POA-6, *P. ostreatus* strain POA-7, *P. ostreatus* strain POA-10 and *P. ostreatus* strain POA-13. Cu content ranged from 15.3 mg/kg to 23.7 mg/kg but below detection in two strains *P. ostreatus* strain POA-6 and *P. ostreatus* strain POA-10. Zn content varied from 189.7 mg/kg to 411.3 mg/kg. Ni content varied from 175 mg/kg to 296.7 mg/kg, Cd content ranged between 57.7 mg/kg and 106.3 mg/kg, Pb content varied from 13.0 mg/kg to 230.7 mg/kg. Cr content ranged between 17.7 mg/kg and 124 mg/kg. All the *Pleurotus* strains showed appreciable levels of antioxidant activities and can be promoted as natural antioxidant preference in food and pharmaceutical

Keywords: 1,1-diphenyl-1-picrylhydrazyl (DPPH) assay, total flavonoids content, total phenolics content, beta carotene content, lycopene content, mineral elements, heavy metals

1. Introduction

Macrofungi from the genus *Pleurotus*, widely known as oyster mushrooms are preferred by many people for their delicate taste, mild yet chewy texture and unique aroma. The world trade of these mushrooms shows an increasing pattern and gives promising opportunity for the traders (Chang 2001). This trend happens not only because of high demand from consumers but also the ability to apply cheap cultivation strategies.

They are found growing naturally on certain rotten woody material (Phillips 2006). They have a wide range of temperature adaptability (Bano & Rajarathnam 1982) and substrate utilization (Peppe 2000). They have been cultivated in large amounts by using lignocelluloses materials such as sawdust, paddy straw, wheat straw and cotton and proven to be successfully cultivated on banana pseudostem, Bahia grass (Siqueira *et al.* 2011), bamboo leaves, lawn grasses (Kumari & Achal 2008), yam peelings (*Dioscorea* sp.), cassava peelings (*Manihot* sp.), wild grass (*Pennisetum* sp.) corn stover (*Zea mays*) and oil palm (*Elaeis guineensis*) fruit fibers (Okhuoya & Okogbo 1991).

Oyster mushrooms (*Pleurotus* species), the third largest commercially produced mushroom in the world (Van 2009) have nutritional and medicinal properties (Garcha *et al.* 1993) Nutritionally, the mushroom has been found to contain vitamins B1 (thiamin), B2 (riboflavin), B5 (niacin), B6 (pyridoxine) and B7 (biotin) (Solomko & Eliseeva 1988). Medically, the species *P. ostreatus* have been reported to decrease cholesterol levels (Hossain *et al.* 2003). The carpophore of the mushroom is also a potential source of lignin and phenol degrading enzymes (Fountoulakis *et al.* 2002). It is also used industrially as a bioremediator (Tsioulpas *et al.* 2002; Barros *et al.* 2007).

In recent times edible mushrooms have not only attracted interest as food but as functional foods. They have also attracted great attention as a source of bioactive metabolites for the development of drugs and nutraceuticals (Terpinc & Abramovic 2010; Orhan & Üstün 2011). Some of them have also been found to be a source of secondary metabolites such as phenolic compounds (Vaz *et al.* 2011), flavonoids, terpenoids, sterols,

ascorbic acid, ergothioneine and carotenoids (García-Lafuente *et al.* 2011). They exhibit high antioxidant properties and they therefore ward off cancers, HIV-1 AIDS and other viral ailments; they are antimutagenic, anti-tumoral and can be used to manage cardiovascular disorders (García-Lafuente *et al.* 2011).

Scientific research has shown that high amounts of antioxidants may prevent oxidative stress caused by free radicals which lead to cell damage, generating of cancer cell and brain cell aging (Wei & Lee 2013). Oxidative stress might occur as a result of either the presence of oxidation agents, decrease in the level of antioxidants or both factors. Under such circumstances, reactive oxygen species (ROS) and free radicals are produced as harmful by-products of oxidation process.

Antioxidants have the ability to scavenge free radicals in the bodies and therefore play an important role in maintaining human health. The human body has its own defense system, such as superoxide dismutase enzyme, glutathione enzyme and catalase, to fight harmful substances and prevent cell damage (Halliwell 1996). However, there is the need to consume foods such as fruits, vegetables and mushrooms which are rich in antioxidants to ensure health security. Therefore, the objectives of this study are to determine the antioxidant activities, total phenolic content and total flavonoids content in the methanolic extract of selected commercial mushroom strains.

2. Materials and Methods

2.1 Materials

2.1.2 Mushroom strains

Twelve *Pleurotus* mushroom strains were used for the study as shown in Table 1. These strains were originally from China, USA and Mauritius and kept in the Mycelium bank of the Mushroom Unit of CSIR-Food Research Institute, Accra, Ghana.

2.2 Methods

2.2.1 Culture maintenance and spawn preparation

All strains were maintained on Potato Dextrose Agar slants and were used to prepare sorghum grain spawn (Oei 1991). Both the cultures and spawn were incubated at 26-28°C until mycelium fully colonized the substrate.

Table 1: Species and strains of oyster mushrooms used in the study.

<i>Pleurotus</i> species/strains	Original Strain (code)	New code	Country of Origin
<i>P. pulmonarius</i>	P35 (AX) (8)	PPA-1	USA
<i>P. pulmonarius</i>	P15 (M53) (9)	PPA-2	USA
<i>P. ostreatus</i>	P6 (012A)	POA-5	USA
<i>P. ostreatus</i>	P47 (HK) (12)	POA-6	USA
<i>P. ostreatus</i>	P25 (A8)	POA-7	USA
<i>P. ostreatus</i>	P31 (JB) (13)	POA-9	USA
<i>P. ostreatus</i>	P8 (Rh) (7)	POA-10	USA
<i>P. ostreatus</i>	P15 (B soy 5)	POA-11	USA
<i>P. ostreatus</i>	P9RL (15)	POA-13	USA
<i>P. citrinopileatus</i>	P096 (14)	PCC-1	CHINA
<i>P. sapidus</i>	P969 (16)	PSC-1	CHINA
<i>P. ostreatus</i> (control)		EM-1	MAURITIUS

2.2.2 Composting of sawdust and mushroom production

Sawdust compost preparation and mushroom production were carried out in accordance to Obodai *et al.* (2007).

2.2.3 Preparation of the sample

Preparation of the mushroom samples was determined according to Tsai *et al.* (2009). The mushrooms were lyophilized (Labconco, Missouri) and ground to obtain fine powder. About 10 g samples were extracted by stirring with 100 ml of ethanol at 25°C at 20×g for 24 hours and filtration through Whatman No. 1 filter paper. The residue was extracted with two additional 100 ml portions of ethanol as described above and combined ethanolic extracts were concentrated under reduced pressure below 40°C to obtain the crude extract. The crude extracts were redissolved in ethanol at concentration 20 mg/ml and stored at 4°C for further analyses.

2.2.4 Determination of total phenolic content

Total phenolic content in extracts was determined by using Folin-Ciocalteu reagent based on method of Harborne (1989) with several modifications. Each sample (150 µl, 10 mg/ml) was added with distilled water (1200 µl) and aqueous sodium carbonate, Na₂CO₃ solution (450 µl). About 100µl of Folin-Ciocalteu reagent was added to the mixture and agitated. The mixture was allowed to stand for 90 minutes and the absorbance was measured at 760 nm by using UV/visible spectrophotometer (SpectraMax Plus384, United States). The concentration of total phenolic compounds was calculated based on standard curve of tannic acid (0.2-1.0 mg/ml) and the results were expressed as µg of gallic acid equivalent (GAE/g) per gram of the extracts which resembled

a linear equation, $y = 0.624x - 0.939$, where $R^2 = 0.995$.

2.2.5 Determination of total flavonoid content

The aluminium chloride colorimetric method as described by Barros *et al.* (2007) was modified and used to determine flavonoid content. Mushroom extract (100 μ l, 10 mg/ml) was mixed with distilled water (500 μ l) and sodium nitrite, NaNO₂ (5%, 30 μ l). The mixture was allowed to stand for 5 minutes. Aluminium chloride solution, AlCl₃.H₂O (10%, 60 μ l) was added to the mixture and left for 6 minutes. Sodium hydroxide, NaOH (1M, 200 μ l) and distilled water (110 μ l) were added to the solution and mixed well. Intensity of the mixed solution was measured at 510 nm (SpectraMax Plus384, United States) and the concentration of total flavonoids contents were calculated as equivalent to standard rutin graph (RE), $y = 0.0101x + 0.2238$, where $R^2 = 0.9563$.

2.2.6 Scavenging 1,1-diphenyl-1-picrylhydrazyl (DPPH) radicals

This assay was performed by following the standard method of Bloise (1958) with several modifications. DPPH solution was prepared by dissolving 5 mg DPPH powder in 2 ml of ethanol, sealed in aluminium foil and kept in a fridge. Accurately 100 μ l of test samples (0.6-20.0 mg/ml) in ethanol was added with 5 μ l DPPH solution in 96-well microtiter plates. Mixture was incubated in the dark for 30 min. Absorbance was measured at 517 nm (SpectraMax Plus384, United States) and IC₅₀ value (concentrations of each sample required to give 50% of the optical density shown by control) was calculated. Inhibition of DPPH free radicals was calculated by using following formula:

$$\text{Inhibition of DPPH radical (\%)} = 100 \times (A_{\text{control}} - A_{\text{sample}}) / A_{\text{control}}$$

Where, A_{control} = Absorbance of the control solution (containing all reagents except the test extract)

A_{sample} = Absorbance of the test extract

All test analyses were run in at least three replicates and averaged. Standard antioxidants such as rutin and DPPH were used as positive control.

2.2.7 Carotenoids

To determine β -carotene and lycopene, methanolic extract of 100 mg of dried mushroom samples was prepared. The extract was vigorously shaken with 10 ml of acetone-hexane mixture (in the ratio of 4:6) for 1 minute and filtered through Whatman No. 4 filter paper. The absorbance of the filtered extract was measured at 453, 505 and 663 nm. Beta carotene and lycopene content were calculated by using the following equations respectively (Barros *et al.* 2007):

For beta carotene: $0.216 \times A_{663} - 0.304 \times A_{505} + 0.452 \times A_{453}$ and

For lycopene: $-0.0458 A_{663} + 0.372 A_{505} - 0.0806 A_{453}$.

The results were expressed as μ g of carotenoid/g of extract.

2.2.8 Qualitative Test for alkaloids

Crude extract of mushroom was mixed with 2ml of 1% HCl and heated gently. Mayer's and Wagner's reagents were then added to the mixture. Turbidity of the resulting precipitate was taken as evidence for the presence of alkaloids.

2.2.9 Determination of minerals and heavy metals

An amount of 0.2 gram was weighed into labeled digestion tubes and dissolved in 2 ml concentrated HNO₃. The solution was heated at 450°C for 4 hours and later dissolved in 1 ml concentrated H₂SO₄, 1 ml HNO₃ and 1 ml H₂O₂, and then diluted with double deionised water up to a volume of 25 ml. A blank digest was carried out by following the above procedure. Contents of minerals and heavy metals in the mushroom samples were determined by using atomic absorption spectrophotometer (Perkin Elmer precisely A Analyst 400)

2.3 Statistical analysis of experimental data

The data obtained was analyzed by GLM procedure of SAS program. LSD was computed to test for significance difference among the mushroom strains at the 5 percent level of significance.

3. Results

3.1 Total phenolic content

The total phenolic content, expressed as μ g of GAEs/g of dry mushroom is shown in Table 2. The highest amount of phenolic compounds was 3.58 μ g of GAEs/g and was recorded by *P. ostreatus* strain POA-7, the lowest amount was 1.48 μ g GAEs/g and was recorded by *P. ostreatus* strain POA-6. Grand mean for the 12 mushroom samples was 2.5 μ g GAEs/g with 0.36 coefficient of variation. Statistical analysis showed significant differences among the 12 strains ($P < 0.001$). *P. sapidus* strain PSC-1 was not significantly different from the control strain, *P. ostreatus* strain EM-1.

Table 2. Mean concentrations ($\mu\text{g/g}$) of total phenols and total flavonoids in 12 cultivated mushroom strains

Strains	Total phenol	Total flavonoids
<i>P. pulmonarius</i> strain PPA-1	2.98	0.63
<i>P. pulmonarius</i> strain PPA-2	2.31	1.80
<i>P. ostreatus</i> strain POA-5	2.95	1.63
<i>P. ostreatus</i> strain POA-6	1.48	0.70
<i>P. ostreatus</i> strain POA-7	3.58	0.67
<i>P. ostreatus</i> strain POA-9	2.39	0.70
<i>P. ostreatus</i> strain POA-10	2.40	0.20
<i>P. ostreatus</i> strain POA-11	2.38	0.50
<i>P. ostreatus</i> strain POA-13	2.38	0.53
<i>P. sapidus</i> strain PSC-1	2.83	2.03
<i>P. citrinopileatus</i> strain PCC-1	2.64	0.57
<i>P. ostreatus</i> strain EM-1	2.88	0.90
Grand mean	2.5	0.85
CV	0.36	9.89
LSD _(0.05)	0.02	0.14

3.2 Total flavonoid content

Total flavonoid contents in the mushroom extracts varied from 0.20 to 2.03 μg of RE/g of dry weight of extracts with a grand mean of 0.85 μg of RE/g of dry weight of extracts and 9.89 coefficient of variation. *P. sapidus* strain PSC-1 recorded the highest content, the lowest was recorded *P. ostreatus* POA-10. Statistically, the test showed significant difference among the 12 mushroom strains ($P < 0.001$)

3.3 Scavenging effect on 2,2-diphenyl-1-picrylhydrazyl (DPPH) radicals

The concentrations of extracts capable of inhibiting 50% of radical solution (IC_{50} value) are presented in Table 3. The extract with the lowest IC_{50} value gave the greatest antioxidant activity. The IC_{50} values varied from 43.21 $\mu\text{g/ml}$ to 52.03 $\mu\text{g/ml}$ for all the mushroom samples tested. On the whole, the inhibition of DPPH radical by *P. sapidus* strain PSC-1 and *P. ostreatus* strain POA-5 exhibited the highest activity.

3.4 Beta carotene and Lycopene contents

Beta carotene and lycopene contents are presented in Table 3. Beta carotene contents in the mushroom extracts varied from 0.60 to 11.46 $\mu\text{g/g}$. *P. ostreatus* strain POA-5 gave the highest content while the lowest was recorded by *P. ostreatus* strain POA-6.

Lycopene contents in the mushroom extracts varied from 0.04 to 4.28 $\mu\text{g/g}$. *P. ostreatus* POA-10 gave the highest content, while *P. ostreatus* strain POA-11 gave the lowest.

In all the 12 mushroom strains Mayer's test for alkaloid proved positive (Table 3)

Table 3. Presence of alkaloid and mean concentrations of IC_{50} , β -carotene, Lycopene for 12 cultivated oyster mushroom strains

Strains	IC_{50} ($\mu\text{g/ml}$)	β -carotene ($\mu\text{g/g}$)	Lycopene ($\mu\text{g/g}$)	Presence of alkaloid
<i>P. pulmonarius</i> strain PPA-1	46.72	3.26	4.08	+
<i>P. pulmonarius</i> strain PPA-2	46.03	5.78	0.22	+
<i>P. ostreatus</i> strain POA-5	43.62	11.46	0.02	+
<i>P. ostreatus</i> strain POA-6	48.30	0.60	1.74	+
<i>P. ostreatus</i> strain POA-7	47.25	6.72	0.05	+
<i>P. ostreatus</i> strain POA-9	52.03	3.15	2.29	+
<i>P. ostreatus</i> strain POA-10	47.98	0.99	4.28	+
<i>P. ostreatus</i> strain POA-11	47.70	4.15	0.04	+
<i>P. ostreatus</i> strain POA-13	49.87	1.60	1.26	+
<i>P. sapidus</i> strain PSC-1	43.21	3.88	0.87	+
<i>P. citrinopileatus</i> strain PCC-1	49.87	1.60	1.26	+
<i>P. ostreatus</i> strain EM-1	47.40	7.65	3.00	+

3.5 Evaluation for mineral elements and heavy metals

The results for mineral elements and heavy metals concentrations are presented in Tables 4 and 5. Mg content varied from 660.0 mg/kg in *P. ostreatus* strain POA-6 to 1993.7 mg/kg in *P. citrinopileatus* strain PCC-1. Fe content ranged from 349.0 mg/kg in *P. pulmonarius* strain PPA-2 to 1374.0 mg/kg in *P. ostreatus* strain POA-9

and could not be detected in *P. ostreatus* strain POA-7. Ca content ranged from 22.0 mg/kg in *P. ostreatus* strain POA-6 to 415.3 mg/kg in *P. citrinopileatus* strain PCC-1. Mn content varied from 10.7 mg/kg in *P. ostreatus* strain POA-11 to 48.3 mg/kg in *P. sapidus* strain PSC-1 but below detection in the six strains *P. pulmonarius* strain PPA-2, *P. ostreatus* strain POA-5, *P. ostreatus* strain POA-6, *P. ostreatus* strain POA-7, *P. ostreatus* strain POA-10 and *P. ostreatus* strain POA-13. Cu content ranged from 15.3 mg/kg in *P. ostreatus* strain POA-13 to 23.7 mg/kg in *P. ostreatus* strain POA-5 but below detection in two strains *P. ostreatus* strain POA-6 and *P. ostreatus* strain POA-10. Zn content varied from 189.7 mg/kg in *P. ostreatus* strain POA-6 to 411.3 mg/kg in *P. sapidus* strain PSC-1.

Statistically there was significant difference among the mushroom strains for heavy metals. Ni content varied from 175 mg/kg in *P. ostreatus* strain POA-13 to 296.7 mg/kg in *P. citrinopileatus* strain PCC-1. Content of Cd ranged from 57.7 mg/kg in *P. ostreatus* strain EM-1 to 106.3 mg/kg in *P. sapidus* strain PSC-1. Pb content ranged between 13.0 mg/kg in *P. ostreatus* strain EM-1 and 230.7 mg/kg in *P. ostreatus* strain POA-13. Cr content ranged from 17.7 in *P. pulmonarius* strain PPA-2 to 124 mg/kg in *P. ostreatus* strain EM-1.

Table 4. Mean concentrations of mineral elements (mg/kg) of *Pleurotus* species

Strains	Mg	Fe	Ca	Mn	Cu	Zn
<i>P. pulmonarius</i> strain PPA-1	1790.0	643.3	100.3	12.3	19.7	318.3
<i>P. pulmonarius</i> strain PPA-2	1128.0	349.0	196.3	nd	17.0	340.3
<i>P. ostreatus</i> strain POA-5	1376.7	478.0	25.7	nd	23.7	389.0
<i>P. ostreatus</i> strain POA-6	660.0	1072.7	22.0	nd	nd	189.7
<i>P. ostreatus</i> strain POA-7	2072.0	nd	256.3	nd	18.0	351.7
<i>P. ostreatus</i> strain POA-9	1686.0	1374.0	246.7	19.3	23.0	393.7
<i>P. ostreatus</i> strain POA-10	1748.3	1072.7	22.0	nd	nd	283.7
<i>P. ostreatus</i> strain POA-11	1924.7	553.3	345.0	10.7	16.3	336.0
<i>P. ostreatus</i> strain POA-13	1327.0	372.3	37.0	nd	15.3	301.7
<i>P. sapidus</i> strain PSC-1	1232.0	553.3	256.3	48.3	17.3	411.3
<i>P. citrinopileatus</i> strain PCC-1	1993.7	1269.7	415.3	19.0	20.0	322.0
<i>P. ostreatus</i> strain EM-1	1555.3	464.0	202.3	12.3	23.3	340.0
Grand mean	1580.7	805.6	218.0	11.5	17.0	308.6
CV	0.65	1.12	0.57	17.98	10.92	12.87
LSD _(0.05)	17.1	15.1	2.1	3.4	3.1	66.4

nd = not detected

Table 5. Mean concentrations of heavy elements (mg/kg) of *Pleurotus* species

Strains	Ni	Cd	Pb	Cr
<i>P. pulmonarius</i> strain PPA-1	217.3	66.0	22.3	66.7
<i>P. pulmonarius</i> strain PPA-2	236.7	86.3	57.3	17.7
<i>P. ostreatus</i> strain POA-5	195.0	84.0	148.0	78.3
<i>P. ostreatus</i> strain POA-6	207.0	91.0	149.0	121.7
<i>P. ostreatus</i> strain POA-7	244.3	84.0	55.7	16.3
<i>P. ostreatus</i> strain POA-9	218.7	70.3	102.0	110.0
<i>P. ostreatus</i> strain POA-10	208.7	91.0	226.0	27.3
<i>P. ostreatus</i> strain POA-11	258.3	72.3	110.7	110.7
<i>P. ostreatus</i> strain POA-13	175.3	77.3	230.7	55.7
<i>P. sapidus</i> strain PSC-1	251.7	106.3	86.0	32.0
<i>P. citrinopileatus</i> strain PCC-1	296.7	107.3	54.7	92.0
<i>P. ostreatus</i> strain EM-1	243.3	57.7	13.0	124.0
Grand mean	229.7	80.4	116.2	75.9
CV	3.64	15.49	32.18	27.1
LSD _(0.05)	14.0	18.1	62.6	34.3

4. Discussion

Mushrooms are a good source of proteins, vitamins and minerals and are low in fat and sugars. The phytochemical analysis conducted on the *Pleurotus* strains showed that they are rich in phenols, flavonoids, carotenoids and mineral elements. These phytochemicals have also been observed in mushrooms by other workers. In the present study, the trend for total phenolic, flavonoids and carotenoids contents differed from one strain to another. This demonstrates the fact that, every strain has its own unique mechanisms in the synthesis

and metabolism of these phytochemicals.

Earlier work by Kim *et al.* (2008) showed that the total phenolic content for both *Pleurotus eryngii* and *Pleurotus ostreatus* were 0.03 mg/g and 0.09 mg/g of dry weight which are higher than the values for the *Pleurotus* strains in the current study. These are also lower than what was observed by Jayakumar *et al.* (2009) for *Pleurotus ostreatus* which gave 0.71 mg/g of dry weight. In another study, Shirmila & Radhamany (2013) recorded total phenolic contents of 5.5 mg/g and total flavonoids of 3.6 mg/g in methanolic extracts of *Macrolepiota mastoidea*, which are higher than those observed in the present study. Also, in a study of nutritional and nutraceutical potential of wild edible Macrolepiotoid mushrooms of North India, Babita & Narender (2014) observed an average of 5.90 mg/g, 11.0 mg/g and 16.0 mg/g total phenols in *Macrolepiota dolichuala*, *M. procera* and *M. rhacodes* respectively; similarly an average of 1.7 mg/g, 1.46 mg/g and 1.30 mg/g of total flavonoid in *Macrolepiota dolichuala*, *M. procera* and *M. rhacodes* respectively.

The total phenol contents in the present study were lower than what they observed, however the total flavonoid contents of three of the *Pleurotus* strains compared favourably with those of the Macrolepiotoid mushrooms. In a study by Arbaayah & Umi Kalsom (2013) on antioxidant properties of oyster mushrooms and split gill mushroom they found total phenolic content in *P. pulmonaris* to be 44.90±0.94 mg of TAE/g dry weight of the extract in the same study they found 43.91±30 mg of TAE/g dry weight of extracts for *P. ostreatus*. For total flavonoids, *P. pulmonaris* gave 9.84±2.93 mg of QE/g dry weight and for *P. ostreatus* 12.97±2.43 mg of QE/g dry weight. These values are higher than the values recorded for these strains in the current study. The difference may be attributed to the substrate used to cultivate the mushrooms and may also be attributed to type of strains used.

Beta carotene contents for the Macrolepiotoid mushrooms were far lower than those of the *Pleurotus* spp in the present study. Similarly with exception of two *Pleurotus* strains, the contents of lycopene in the *Pleurotus* mushrooms were higher than those in Macrolepiotoid mushrooms.

Analysis of carotenoids in *Macrolepiota* species by Babita & Narender (2014) showed that contents for beta carotene and lycopene ranged from 0.12 - 0.29 µg/g and 0.05 - 0.12 µg/g respectively, the values obtained in the current study are higher than these values. The range for the contents of beta carotene and lycopene in the present work are 0.6 - 11.46 µg/g and 0.02 - 4.28 µg/g, respectively. Their observations for beta carotene were as follows 0.12 µg/g, 0.29 µg/g and 0.20 µg/g for *Macrolepiota dolichuala*, *M. procera* and *M. rhacodes* respectively, whilst that for lycopene were: 0.05 µg/g, 0.07 µg/g and 0.12 µg/g for *Macrolepiota dolichuala*, *M. procera* and *M. rhacodes* respectively.

Previous studies on inhibition of DPPH radical by Arbaayah & UmiKalsom (2013) showed that *P. pulmonariis* and *P. ostreatus* gave IC₅₀ values of 5.61 mg/ml, 5.61 mg/ml and 8.88 mg/ml, respectively. In another work by Menaga *et al.* (2013), methanol extract of *P. florida* mushroom showed DPPH radical scavenging IC₅₀ value of 50 µg/ml. These values are lower than what were observed in the current work.

Of the mineral elements investigated *P. sapidus* strain PSC-1 had the highest value of Ca, Mn and Zn. It has been reported that mushrooms possess very effective mechanisms that enable them to readily absorb heavy metals from their substrates (Turkekul *et al.* 2004). High concentrations of heavy metals were observed in the fruiting bodies of *Pleurotus sajor-caju* collected adjacent to heavy metal smelters and oil polluted areas (Pandiarajan *et al.*, 2012). In the current work the contents for the heavy metals analyzed were higher than those extracted by Pelkonen *et al.* (2006) in fresh *Macrolepiota procera*: 0.059 (Cd), 0.215(Pb) 0.113(As) 0.121 (Ni) mg/Kg.

The heavy metals in these samples may be accounted for by their levels in the biological materials used for the substrate. The results of the present study show that the methanolic extracts of the mushroom strains had significant antioxidant potential that may reveal their therapeutic potentials for several diseases.

5. Conclusion

The results obtained from this study suggest that the methanolic extract of the 12 *Pleurotus* mushroom strains have significant antioxidant activity. They could therefore serve as a rich source of natural rich antioxidant food for the enhancement of the immune system against oxidative damage. They may also have the therapeutic potentials for several diseases and may have applications in food, cosmetics and pharmaceutical industries.

Acknowledgements

The authors are very grateful to the Office of Research and Innovative Division of the University of Ghana, Legon for providing funds for the research. We also wish to acknowledge the technical support offered by the following persons: Mr. Prince Owusu of the Ecological Laboratory University of Ghana; K. A. Obeng, Mr. M. Kwarteng and Mr E. D. Maison.

References

Arbaayah, H.H. & Umi Kalsom, Y. (2013), Antioxidant properties in the oyster mushrooms (*Pleurotus* spp.)

- and split gill mushroom (*Schizophyllum commune*) ethanolic extracts. *Mycosphere*, 4 (4), 661–673.
- Babita, K. & Narender, S. A. (2014), Nutritional and nutraceutical potential of wild edible Macrolepiotoid mushrooms of North India. *International Journal of Pharmacy and Pharmaceutical Sciences*, 6 (2), 200-204.
- Bano, Z. & Rajarathnam, S. (1982), *Pleurotus* Mushroom as a Nutritious Food, In: Chang, S. T. & Quimio, T. H. (eds). *Tropical Mushrooms: Biological Nature and Cultivation methods*. The Chinese University Press.
- Bloise, M.S. (1958), Antioxidant determination by the use of stable free radicals. *Nature*, 181, 1199–2000.
- Barros, L., Ferreira, M.J., Queiros, B., Ferreira, I. & Baptista, P. (2007), Total phenols, ascorbic acid, beta-carotene and lycopene in Portuguese wild edible mushrooms and their antioxidant activities. *Food Chemistry*, 103 (2), 413-419.
- Chang, S.T. (2001), *Mushrooms and mushroom cultivation*. Chichester: John Wiley & Sons Ltd.
- Fountoulakis, M.S., Dokianakis, S.N., Kornaros, M.E., Aggelis, G.G. & Lyberatos, G. (2002), Removal of phenolics in olive mill wastewaters using the white-rot fungus *Pleurotus ostreatus*. *Water Research*, 36, 4735-4744.
- Garcha, H.S., Khann, P.K. & Soni, G.L. (1993), Nutritional importance of mushroom. In: Chang, S.T., Buswell, J.A. & Chiu, S. (Eds.), *Mushroom biology and Mushroom Products*, The Chinese University Press, pp. 227-235.
- García-Lafuente, A., Moro, C., Villares, A., Guillamón, E., Rostagno, M.A., D'Arrigo, M. & Martínez, J.A. (2011), Mushrooms as a source of anti-inflammatory agents. *American Journal of Community Psychology* 48(1-2), 125-141.
- Halliwell, B. (1996), Antioxidants in human health and disease. *Annual Review of Nutrition*, 16, 33–50.
- Harborne, J.B. (1989), General procedures and measurement of total phenolics. *Methods in Plant Biochemistry*, 1, 1–28. London: Academic Press.
- Hossain, S., Hashimoto, M., Choudhury, E., Alam, N., Hussain, S., Hasan, M., Choudhury, S. & Mahmud, I. (2003), Dietary mushroom (*Pleurotus ostreatus*) ameliorates atherogenic lipid in hypercholesterolaemic rats. *Clinical and Experimental Pharmacology and Physiology*, 30, 470-476.
- Jayakumar, T., Thomas, P.A. & Geraldine, P. (2009), In-vitro antioxidant activities of an ethanolic extract of the oyster mushroom, *Pleurotus ostreatus*. *Innovative Food Science and Emerging Technologies*, 10(2), 228–234.
- Kim, M.K., Math, R.K., Cho, K.M., Shin, K. J., Kim, J.O., Ryu, J.S. & Lee, Y.H. (2008), Effect of *Pseudomonas* sp. P7014 on the growth of edible mushroom *Pleurotus eryngii* in bottle culture for commercial production. *Bioresource Technology*, 99 (8), 3306–8.
- Kumari, D. & Achal, V. (2008), Effect of different substrates on the production and non-enzymatic antioxidant activity of *Pleurotus ostreatus* (Oyster mushroom). *Life Sciences Journal*, 5(3), 73–76.
- Siqueira, F.G., Martos, E.T., Silva, R. & Dias, E.S. (2011), Cultivation of *Pleurotus sajor-caju* on banana stalk and Bahia grass based substrates”, *Horticultura Brasileira*, 29, 199–204.
- Menaga, D., Rajakumar, S. & Ayyasamy, P. M. (2013), Free Radical Scavenging Activity of Methanolic Extract Of *Pleurotus Florida* mushroom. *International Journal of Pharmacy and Pharmaceutical Sciences*, 5 (4), 601-606.
- Obodai, M., Dzomeku, M., Awotwe, B., Takli, R.K. & Narh, D. (2007), *Manual on mushroom cultivation technology in Ghana*. CSIR-FRI Technical Report
- Oei, P. (1991), *Manual on mushroom cultivation: techniques, species and opportunities for commercial application in developing countries*. CTA, Wageningen, The Netherlands
- Okhuoya, J.A. & Okogbo, F.O. (1991), Cultivation of *Pleurotus tuber-regium* (Fr) Sing on Various Farm Wastes. *Proceedings of the Oklahoma Academy of Science* 3, 1–3.
- Orhan, I. & Üstün, O. (2011), Determination of total phenol content, antioxidant activity and acetylcholinesterase inhibition in selected mushrooms from Turkey. *Journal of Food Composition and Analysis*, 24, 386–390.
- Pandiarajan, G., Govindaraj, R., Mareeswaran, J. & Kumari, B.M. (2012), Antibacterial activity and heavy metal accumulation of edible oyster mushroom (*Pleurotus sajor-caju*) grown on two substrates, *International Journal of Pharmacy and Pharmaceutical Sciences*, 4 (2), 238–240.
- Phillips, R. (2006), *Mushrooms*. McMillan, p. 266
- Poppe, J. (2000), Use of agricultural waste materials in the cultivation of mushrooms. *Mushroom Science*, 15, 3-23.
- Shirmila, J.G. & Radhamany, P.M. (2013), Invitro antioxidant activities, total phenolics and flavonoid of wild edible mushroom *Macrolepiota mastoidea* (fr.) Singer. *International Journal of Pharmacy and Pharmaceutical Sciences*, 5 (2), 161-166.
- Solomko, E.F. & Eliseeva, G.S. (1988), Biosynthesis of vitamins B by the fungus *Pleurotus ostreatus* in a submerged culture. *Prikl Biokhim Microbiol*, 24, 164-169.
- Tsai, S.Y., Huang, S.J., Lo, S.H., Wu, T.P., Lian, P.Y. & Mau, J.L. (2009), Flavour components and antioxidant properties of several cultivated mushrooms. *Food Chemistry*, 113(2), 578–584.
- Terpinic, P. & Abramovic, H. (2010), A kinetic approach for evaluation of the antioxidant activity of selected

phenolic acids. *Food Chemistry*, 121, 366–371.

Tsioulpas, A., Dimou, D., Iconomou, D. & Aggelis, G. (2002), Phenolic removal in olive oil mill wastewater by strains of *Pleurotus spp.* in respect to their phenol oxidase (laccase) activity. *Bioresource Technology*, 84, 251-257.

Turkekul, I., Elmastas, M. & Tuzen, M. (2004), Determination of iron, copper, manganese, zinc, lead and cadmium in mushroom samples from Tokat, Turkey. *Food Chemistry*, 84, 389-392.

Van, N. (2009), Developments in the European Mushroom Industry. Presented at 2nd African Conference on Edible and Medicinal mushrooms, Accra, Ghana.

Vaz, A. J., Barros, L., Martins, A., Santos-Buelga, C., Vasconcelos, M. H., & Ferreira, I. C. F. R. (2011), Chemical composition of wild edible mushrooms and antioxidant properties of their water soluble polysaccharidic and ethanolic fractions. *Journal of Food Composition and Analysis*, 126, 610–616.

Wei, Y. & Lee, H. (2013), Oxidative Stress, Mitochondrial DNA Mutation, and Impairment of Antioxidant Enzymes in Aging, *Experimental Biology and Medicine*, 227, 671–682.

The IISTE is a pioneer in the Open-Access hosting service and academic event management. The aim of the firm is Accelerating Global Knowledge Sharing.

More information about the firm can be found on the homepage:
<http://www.iiste.org>

CALL FOR JOURNAL PAPERS

There are more than 30 peer-reviewed academic journals hosted under the hosting platform.

Prospective authors of journals can find the submission instruction on the following page: <http://www.iiste.org/journals/> All the journals articles are available online to the readers all over the world without financial, legal, or technical barriers other than those inseparable from gaining access to the internet itself. Paper version of the journals is also available upon request of readers and authors.

MORE RESOURCES

Book publication information: <http://www.iiste.org/book/>

IISTE Knowledge Sharing Partners

EBSCO, Index Copernicus, Ulrich's Periodicals Directory, JournalTOCS, PKP Open Archives Harvester, Bielefeld Academic Search Engine, Elektronische Zeitschriftenbibliothek EZB, Open J-Gate, OCLC WorldCat, Universe Digital Library, NewJour, Google Scholar

