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***J. Nat. Prod. Plant Resour.*, 2013, 3 (6):29-33**
(http://scholarsresearchlibrary.com/archive.html)

Journal of Natural Product and Plant Resource
Scholars Research Library

ISSN : 2231 – 3184
CODEN (USA): JNPPB7

Proximate composition and anti-nutritive factors in some wild edible medicinal macrofungi

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ABSTRACT

Fruiting bodies of eight (8) wild edible macro-fungi (Agaricus campestris, A. silvaticus, Coprinus comatus, C. atramentarius, Pleurotus ulmarius, Bondazewia berkeleyi, Auricularia auricula and Ganoderma lucidum) were analyzed for their nutrients and anti-nutrients content. The result showed that these mushrooms contained appreciable amount of crude protein (5.48-26.78%), carbohydrate (37.14-83.99%), crude fibre (2.13-21.0%), moisture (3.0-10.55%); ash (3.2-25.1%) and energy value (208.05-377.68 kcal/100g). The concentration of crude lipid (0.23-3.30%) and fatty acids (0.18-2.64%) was low, hence valuable nutritional ingredient. The anti-nutrients, tannins (0.10-0.571 mg/g), phytates (0.115-0.453 mg/g), soluble oxalates (0.264-1.06 mg/g), insoluble oxalates (0.186-0.528 mg/g) and cyanide content (0.09-0.37 mg/g) were also relatively low, hence below the permissible toxic levels. Therefore, these medicinal mushrooms are a rich source of proteins, fibers and carbohydrates, and are potential sources of nutraceuticals. They could be useful for those who suffer from anaemia and hyperlipidemia since they are very low in lipid.

Key words: Macrofungi, proximate composition, anti-nutrients, nutraceutical

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INTRODUCTION

Wild edible mushrooms are traditionally used by many African countries as food and medicine; they are appreciated not only for texture and flavor but also for their chemical and nutritional characteristics [1-2]. Mushrooms are valuable healthy foods, low in calories, fats, and essentials fatty acids, and high in vegetable proteins, vitamins and minerals [3-4]. These mushrooms have a worldwide distribution and are not only sources of nutrients but also have been reported as therapeutic foods, useful in preventing diseases such as hypertension, hypercholesterolemia and cancer [5-7]; they are utilized as dried whole, powder or capsules and as tablets for promoting health in humans [8]. These functional characteristics are mainly due to the presence of dietary fiber and in particular chitin and beta glucans [9]. Each mushroom type produces a specific set of metabolites capable of dealing with the set of microbes that coexist in that specific environment [10].In China and Japan, *Ganoderma lucidum* are cultivated and utilized as source of feed supplement [11-12].

Anti-nutrients are natural or synthetic compounds that interfere with the absorption of nutrients. Phytic acid has a strong binding affinity to minerals such as calcuim, magnesium, iron, copper and zinc. This results in precipitation, making the minerals unavailable for absorption in the intestines [13-14]. Oxalates bind to calcium and prevent its

absorption in the human body. Approximately 75% of all kidney stones are composed primarily of calcium oxalate [15], with hyperoxaluria considered to be a primary risk factor for this type of stones [16]. Although it was reported that dietary oxalate contributes no more than 10–20% of the oxalate excreted in urine under normal conditions [15], more recent work [17-18] suggested that even in the absence of gastrointestinal disorders, intestinal absorption of dietary oxalate can make a more considerable contribution to urinary oxalate output. Another particular widespread form of anti-nutrients is the flavanoids which are a group of polyphenolic compounds that include tannins. These compounds chelate metals such as iron and zinc and reduce the absorption of these nutrients, but they also inhibit digestive enzymes and may also precipitate proteins [19]. These anti-nutritional factors must be inactivated or removed, if values of food substances are to be fully maintained. There are reports on cultivated and wild edible mushrooms and their nutritional value from some regions of the world [4, 20], however, there has been paucity of information on these tropical wild edible species from Nigeria. The justification informs this study whose aim is to determine the nutrients and anti-nutrients profiles of *A. campestris, A. silvaticus, C. comatus, C. atramentarius, P. ulmarius, B. berkeleyi, A. auricula* and *G. lucidum* in order to make deductions for general consumption on their food value as well as their species differences as food items.

MATERIALS AND METHODS

Samples collection and treatment

The fresh sporocarps of eight mature mushroom species (*A. campestris, A. silvaticus, C. comatus, C. atramentarius, P. ulmarius, B. berkeleyi A. auricula* and *G. lucidum*) were collected from the field in the rainy season (September to October) within the University of Uyo campus and its environs. Specimen identifications and authentication were done by Dr. J. Essien and the voucher specimens were deposited in the School of Pharmacy herbarium, University of Uyo, Nigeria. The macroscopic descriptions, including size, shape, color, texture, and odor, were noted. The color of the carpophore, shape of the cap and stipe, color of the flesh and latex, and its smell and habitat were also noted. The mushroom samples were packed in opaque plastic bags, which were stored in a refrigerator during transportation to analytical laboratory. The samples were carefully cleaned manually to remove any extraneous materials, cut, sun-dried and oven-dried (Gallenkamp, DV 333) at 45°C for 40 h to constant weight. Dried samples were pulverized using an agate homogenizer, and stored in pre-cleaned polyethylene bottles, prior to analyses. All reagents were of analytical grade, except otherwise stated.

Proximate compositional analysis

Moisture content was obtained from fresh materials; total lipid, protein, ash, crude fibre and carbohydrate were determined from oven-dried powder using standard procedures. The moisture content was obtained by drying in a moisture determination apparatus (Precisa HA60) at 110 ºC until circulation was complete; ash, from the incinerated residue obtained at 550°C after 3 h; crude protein, by the Kjeldahl method with a conversion factor of 6.25 [21-22]; crude fat, gravimetrically determined after Soxhlet extraction with petroleum ether [23]. The crude fat was converted into fatty acids by multiplying with conversion factor of 0.8 [24-25]. The total carbohydrate was calculated as 100% - (% moisture+ % ash+ % crude protein+ % fat+ % fibre) [22, 26]. Total energy values were calculated by multiplying the amounts of protein and carbohydrate by the factor of 4 kcal/g and lipid by the factor of 9 kcal/g [21, 27]. Data points represent mean of three determinations and proximate values were reported in percentage.

Determination of anti-nutritional factors

Phytate determination

Extraction and precipitation of phytate were done through phytic acid determination using the procedure described by Lucas and Markaka [28]. This entails the weighing of sample (2g) into a 250 mL conical flask. 2% conc. HCl (100 mL) was used to soak the samples in the conical flask for 3 h and then filtered through a double layer filter paper. Sample filtrate (50 mL) was placed in a 250 mL beaker and distilled water (107 mL) added to give/ improve proper acidity. 0.3% ammoniumthiocyanate solution (10 mL) was added to each sample solution as indicator and titrated with standard iron chloride solution which contained 0.00195 g iron/mL and the end point was signified by brownish-yellow colouration that persisted for 5 min. The percentage phytic acid was calculated.

Tannins determination

Tannin values were obtained by adopting the method of Jaffe [29]. Each sample (1g) was dissolved in distilled water (10 mL) and agitated, left to stand for 30 min. at room temperature. The samples were centrifuged and the extracts recovered; the supernatant (2.5 mL each) were dispersed into 50 mL volumetric flask. Similarly, standard tannic

acid solution (2.5 mL) was dispersed into separate 50 mL flasks. Folin-dennis reagent (1.0 mL) was measured into each flask followed by the addition of saturated Na_2CO_3 solution (2.5 mL). The mixture was diluted to 50 mL in the flask and incubated for 90 min at room temperature. The absorbance of each sample was measured at 250 nm with the reagent blank at zero. The % tannin was calculated.

Cyanogenic glycoside determination

The method used was alkaline picrate method of Onwuka [30]. The samples (5 g each) in conical flasks were added distilled water (50 mL) and allowed to stand overnight. Alkaline picrate (4 mL) was added to sample filtrate (1 mL) in a corked test tube and incubated in a water bath for 5 min. A colour change from yellow to reddish brown after incubation for 5 min in a water bath indicated the presence of cyanides. The absorbance of the samples was taken at 490 nm and that of a blank containing distilled water (1 mL) and alkaline picrate solution (4 mL) before the preparation of cyanide standard curve.

Oxalates determination

The oxalates content of the samples was determined using titration method. The samples (2 g each) were placed in a 250 mL volumetric flask suspended in distilled water (190 mL) for soluble oxalate determination; 6 M HCl solution (190 mL) was added to each of the samples (2 g each) for total oxalate determination. The suspensions were digested at 100ºC for 1h. The samples were then cooled and made up to 250 mL mark of the flask. The samples were filtered, triplicate portions of the filtrate (50 mL) were measured into beaker and four drops of methyl red indicator was added, followed by the addition of concentrated NH_4OH solution (drop wise) until the solution changed from pink to yellow colour. Each portion was then heated to 90ºC, cooled and filtered to remove the precipitate containing ferrous ion. The filtrates were again heated to 90ºC and 5% $CaCl_2$ (10 mL) solution was added to each of the samples with consistent stirring. After cooling, the samples were left overnight. The solutions were then centrifuged at 2500 rpm for 5 min. The supernatant were decanted and the precipitates completely dissolved in 20% H_2SO_4 (10 mL). The total filtrates resulting from digestion of the samples (2 g each) were made up to 200 mL. Aliquots of the filtrate (125 mL) was heated until near boiling and then titrated against 0.05 M standardized $KMnO_4$ solution to a pink colour which persisted for 30 sec. The oxalate contents of each sample were calculated. Insoluble oxalate, presumed to be primarily calcium oxalate, was computed as the difference between total and soluble oxalate [31]. All determinations were performed in triplicates and presented in mg/g.

RESULTS AND DISCUSSION

Proximate composition

The chemical composition and calculated energy values for the studied mushroom species are shown in Table 1. It is known that the moisture content of mushrooms depends on their harvesting time, maturation period and environmental conditions such as humidity and temperature in growing period, and storage conditions [3]. The moisture content of all studied mushroom species ranged from 3.00% to 10.55%. The moisture contents of these mushrooms were similar to other reports for some edible mushrooms [12, 32]. The ash contents were observed between 3.20% and 25.10% which indicates that the mushroom contains some nutritionally important minerals. Mattila *et al*. [33] reported that the main constituents in the mushroom ash were K and P (approximately 60%).

The major compounds of mushrooms are proteins and carbohydrates. It is reported that the protein contents of mushrooms are affected by a number of factors, such as the type of mushrooms, the stage of development, the part sampled, level of nitrogen available and the location [34]. Total protein content, varying between 21-50%, can be accepted as high when compared with meat, milk, egg and fish [22] and some other mushroom species [4]. In this study, the highest protein content was found for *A. silvaticus* (26.78%) and the lowest was *G. lucidum* (5.48%). Our findings are in consonance with published data for *Pleuroteus* species (5.30%, 6.56%), *A. auricula*-judea (5.52%) [35] and *G. lucidum* (16.79%) [12].

Mushroom carbohydrates include glucans, mono- and disaccharides, sugar alcohol, glycogen and chitin (Kurztman, 1997). The results showed that the fruiting bodies are a good source of carbohydrates (37.14-83.99%). The carbohydrate content of the mushrooms appeared similar to previous reports for *Pleuroteus* species (61.24 & 69.93%), *A. auricula* (77.74%) [35] and *G. lucidum* (63.27%) [12]. The carbohydrate contents of some wild edible mushrooms from nitrogen free extracts were found to be between 41.00% and 65.00% [23]. The crude fibre (2.13-21.0%) was proximate with values for *Pleuroteus* species, *A. auricula*, and *G. lucidum* [12, 35]. Calculated energy values of edible wild mushrooms varied from 208.05 to 377.68 kcal/100 g in dry matter basis (Table 1).

Lipid compounds such as free fatty acids, tri-, di- and monoglycerides, phospholipids, sterols and derivatives can be extracted from mushrooms as crude fat [3]. The crude lipid (0.23-3.30%) and fatty acids (0.18-2.64%) were low (Table 1), hence these mushrooms are a valuable nutritionally healthy ingredient. Low fat food reduces cholesterol level [37]. In the previous reports, it is possible to see various fat contents from 0.80% to 27.50% in dry mushrooms [4, 38].

Table 1: Proximate composition of some wild edible mushrooms

Sample	Moisture content (%)	Ash (%)	Crude protein (%)	Crude fibre (%)	Crude lipid (%)	Fatty acids (%)	Carbohy-drate (%)	Energy value (Kcal/100g)
A. campestris	5.90±0.11	20.20±0.1	12.99±0.12	13.47±0.04	1.0±0.10	0.80±0.11	46.44±0.05	246.72±0.10
A. silvaticus	10.55±0.02	9.03±0.21	26.78±0.17	6.20±0.20	2.15±0.02	1.72±0.01	45.29±0.25	307.63±0.50
C. comatus	8.55±0.01	8.60±1.1	5.51±0.11	5.73±0.50	3.30±0.00	2.64±0.13	68.31±0.65	324.98±0.11
C. atramentarius	8.20±1.0.	21.50±0.00	8.79±0.02	21.00±0.10	2.17±0.01	1.74±0.02	38.34±0.10	208.05±0.15
P. ulmarius	5.50±0.11	25.10±0.1	26.51±0.02	3.40±0.12	2.35±0.01	1.88±0.30	37.14±0.13	275.75±0.03
B. berkeleyi	10.20±0.05	3.90±0.15	6.17±0.15	2.60±0.11	0.23±0.00	0.18±0.09	76.90±0.10	334.35±0.01
A. auricular	7.80±0.12	3.60±0.02	7.30±0.13	4.73±0.10	2.50±0.01	2.00±0.10	74.07±0.12	347.98±0.02
G. lucidum	3.00±0.15	3.20±0.40	5.48±0.11	2.13±0.20	2.20±0.00	1.76±0.05	83.99±0.50	377.68±0.01

Data are mean ± standard deviation (SD) of triplicate results

Anti-nutrients analysis

The anti-nutrients analysis revealed the presence of tannins (0.1-0.571 mg/g), phytates (0.115-0.453 mg/g), and total oxalates (0.680-1.584 mg/g). The cyanide content of the mushrooms was low (0.09-0.37 mg/g) (Table 2). Many authors report soluble and insoluble oxalate as separately measurable components of the oxalate content of foods [39-40]. In food, oxalic acid is typically found as either sodium or potassium oxalate, which are water soluble, or calcium oxalate, which is insoluble. Magnesium oxalate is also poorly soluble in water, although the contribution of this salt to the insoluble fraction of oxalate in food is unclear. The propensity of a specific food to raise urinary oxalate is dependent both on oxalate content and efficiency of absorption, because it is well established that little oxalate catabolism occurs after absorption and >90% of absorbed oxalate can be recovered in the urine within 24–36 h [41]. Furthermore, cyanide taken in the diet is detoxified in the body, resulting in the production of thiocyanate. Thiocyanate has the same molecular size as iodine and interferes with iodine uptake by the thyroid gland [42]. Therefore, some of these anti-nutrients (phytate, oxalate and tannins) can be reduced by proper processing of food [24]. The levels of anti-nutrients in the studied mushrooms were below the permissible toxic levels [43].

Table 2: Anti-nutrients composition of some wild edible mushrooms

Sample	Phytate (mg/g)	Tannins (mg/g)	Cyanide (mg/g)	Soluble oxalate (mg/g)	Insoluble oxalate (mg/g)	Total oxalates (mg/g)
A. campestris	0.283±0.07	0.100±1.00	0.178±0.30	0.968±0.10	0.440±0.10	1.410±0.10
A. silvaticus	0.115±0.01	0.126±0.10	0.090±0.11	0.704±0.12	0.440±0.20	1.144±0.11
C. comatus	0.453±0.10	0.290±0.10	0.239±0.20	0.968±0.10	0.528±0.11	1.496±0.10
C. atramentarius	0.419±0.01	0.571±0.15	0.370±0.30	1.060±0.05	0.500±0.10	1.584±0.13
P. ulmarius	0.321±0.07	0.357±0.50	0.271±0.33	0.880±0.10	0.440±0.15	1.320±0.10
B. berkeleyi	0.269±0.10	0.422±0.10	0.203±0.10	0.704±0.03	0.352±0.05	1.056±0.11
A. auricular	0.183±0.32	0.242±0.45	0.271±0.60	0.792±0.01	0.186±0.13	0.978±0.10
G. lucidum	0.253±0.60	0.126±0.71	0.257±0.40	0.264±0.88	0.410±0.10	0.680±0.10

Data are mean ± standard deviation (SD) of triplicate results.

CONCLUSION

These results suggest that the studied mushrooms are good nutrition sources for mankind who search for new and alternative food and nutrition options. The mushrooms are low in lipid and contain relatively high amount of protein, ash, fibre and carbohydrates. They could be very useful for vegetarians and those who suffer from anaemia and hyperlipidemia; and may be helpful in the reduction of cholesterol, very low-density lipoprotein (VLDL), low-density lipoprotein (LDL), and high-density lipoprotein (HDL).

REFERENCES

[1] P Manzi; L Gambelli; S Marconi; V Vivanti; L Pizzoferrato. *Food Chem.*, **1999**, 65, 477-482.
[2] IO Ezeibekwe; CI Ogbonnaya; CIN Unamba; Osuala, O.M. *Report and Opinion*, **2009**, 1(*4*), 32-36.

[3] EV Crisan; A Sands. In "The Biology and Cultivation of Edible Mushrooms", ST Chang; WA Hayes, Eds., Academic Press, New York, **1978**; pp. 137-168.
[4] D Agrahar-Murugkar; G Subbulakshmi. *Food Chem*., **2005**, 89, 599-603.
[5] P Bobek; L Ozdyn; L Kuniak. *Nahrung*, **1995**, 39, 98.
[6] V Kupin. Proceeding of the 6th International Symposium on *Ganoderma lucidum*. Seoul, II Yang, **1994**; pp. 36-37.
[7] Y Kabir; S Kimura. *J. Nutr. Sci. Vitaminol*., **1989**, 35, 191-194.
[8] P Oei. Mushroom Cultivation. In Appropriate Technology for the Mushroom Growers; CTA, 3rd ed., Backhuys Publishers, Leiden, The Netherlands, **2003**; pp.1-7.
[9] P Manzi; A Aguzzi; L Pizzoferrato. *Food Chem*., **2001**, 73, 321.
[10] Annonymous. URL-1, Medicinal mushrooms a light in the dark. 2005-06-11: htm http://www. Nutraceuticalsworld.com/guide1.htm, **2005**.
[11] S Wachtel-Galor; B Tomlinson; IF Benzie. *British J. Nutr*. **2004**, 91(*2*), 263-269.
[12] AO Ogbe; AD Obeka. *Iranian J. Appl. Ani. Sci*., **2013**, 3 (*1*), 161-166.
[13] P Ekholm; L Virkki; M Ylinen; L Johansson. *Food Chem*., **2003**, 80(*2*): 165–70.
[14] M Cheryan; J Rackis. *Crit Rev Food Sci. Nutr*., **1980**, 13(*4*): 297–335.
[15] HE Williams; TR Wandzilak. *J Urol*. **1989**, 141,742–747
[16] WG Robertson, H Hughe. *Scanning Microsc*., **1993**, 7, 391–402.
[17] M Liebman; W Chai. *Am. J. Clin. Nutr*., **1997**, 65, 1453–1459.
[18] RP Holmes; HO Goodman; DG Assimos. *Kidney Int*., **2001**, 59, 270-276.
[19] GR Beecher. *J. Nutr*., **2003**, 133 (*10*): 3248S-3254S.
[20] L Barros; P Baptista; DM Correia, S Casal, B Oliveira, ICFR Ferreira. *Food Chem*., **2007**, 105, 140-145.
[21] Annonymous."Official Methods of Analysis", Arlington, VA, USA: Association of Official Analytical Chemist (AOAC), 16th ed., **1995**; p. 331.
[22] S Güner; B Dincer; N Alemdag; A Colak; M Tüfekci. *J. Sci. Food Agr*., **1998**, 78, 337-342.
[23] R Sanmee; B Dell; P Lumyong; K Izumori, S Lumyong. *Food Chem*., **2003**, 84: 527-532.
[24] RO Akinyeye; A Oluwadunsin; A Omoyeni. *Elect. J. Environ. Agric. Food Chem.,* **2011**, 10(*1*), 1848-1857.
[25] H Greenfield; DA Southgate. Food Composition Data. Production Management and Use, 2nd Ed. Rome, FAO, **2003**.
[26] P Mattila; P Salo-Vaananen; K Könkö; H Aro; T Jalava. *J. Agr. Food Chem*., **2002**, 50, 6419-6422.
[27] M Fatima; M Al-Subhi. *J. Am. Sci*., **2013**, 9(*5*), 250-255.
[28] GM Lucas; P Markaka. *J. Agric. Ed. Chem.*, **1975**, 23, 13-15.
[29] CS Jaffe. Analytical Chemistry of Food, Vol. 1, Blackie Academic and Professional, New York, **2003**; p. 200.
[30] G Onwuka. Food Analysis and Instrumentation, 3rd ed., Naphohla Prints, A Division of HG Support Nigeria Ltd., **2005**; pp. 133-161.
[31] AB Munro; WA Bassiro. *J. Biol. Appl. Chem.*, **2000**, 12(*1*), 14-18.
[32] AO Ketiku, L Ola. *Trop. Anim. Prod*., **1999**, 2, 169-174.
[33] P Mattila; K Könkö; M Eurola; JM Pihlava; J Astola; L Vahteristo; V Hietaniemi; J Kumpulainen; M Valtonen; V Piironen. *J. Agr. Food Chem.*, **2001**, 49, 2343-2348.
[34] PB Flegg; G Maw. *Mushroom J*., **1977**, 48: 395-403.
[35] CK Shin; CF Yee; LJ Shya; A Atong. *J. Appl. Sci*., **2007**, 7(*15*): 2216-2221.
[36] RH Kurztman. *Mycoscience*, **1997**, 38, 247-253.
[37] MN Gordon; M Kessel. Perspective in Nutrition. McGraw Hill Company, 5th ed., Ohio, New York, **2002**.
[38] BH Huang; KH Yung; ST Chang. *Mushroom Sci*., **1989**, 12, 533-540.
[39] R Hönow; A Hesse. *Food Chem.* **2002**, 78, 511-521.
[40] GP Savage; L Vanhanen; SM Mason; AB Ross. *J. Food Comp. Anal*. **2000**, 13, 201-206.
[41] TD Elder; JB Wyngaarden. *J. Clin. Invest*., **1960**, 39, 1337–1344.
[42] P Bourdoux; F Delange; M Gerard; M Mafuta; A Hudson; MA Ermans. *J. Clin. Endocrinol. Metab*., **1978**, 4b, 613-621.
[43] G Birgitta; C Gullick. Exploring thePotential of Indigenous Wild Food Plants in Southern Sudan. Proceeding of a workshop held in Lokichoggio, Kenya, **2000**; pp. 22-25.