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Proximate and Amino Acid Composition of Wild and Cultivated Edible Mushrooms Collected from Ethiopia

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Abstract: First four cultivated (P.ostreatus, L.edodes, A.bisporus#1, A. bisporus#2) and eight wild (A.campestris, L.sulphureus, T.clypeatus, T.microcarpus#1, T.aurantiacus, T.microcarpus#2, T.letestui and Termitomyces spps) edible mushrooms collected from Ethiopia were analyzed for their proximate composition. Then to measure the quality of the crude protein, nine mushrooms were selected for further investigation of their amino acid composition. The proximate (g/100g) composition in dry weight basis was significantly ($P<0.05$) varied and ranged: protein 6.84-36.7, fat 1.34-5.16, ash 1.75 - 25.3 (T.microcarpus#2), dietary fiber 6.40-13.4, utilizable carbohydrate 32.3-82.3 and energy 266.8 -381.1 kcal on average. All mushrooms contained 18 amino acids ranging in percentage (%): Asp (4.87-10.4), Glu (0.64-2.75), Ser (1.46-2.64), Asn (1.90-4.45), Gln (1.06-4.51), His-Gly-Thr (4.05-15.9), Ala (1.07-6.6), Arg (1.62-6.77), Tyr (1.176-10.0), Cys-SS-Cys (1.08-3.67), Val-Met (2.27-6.15), Phe (0.93-1.75), Ile (0.84-1.71), Leu (0.79-9.66) and Lys (0.869-2.37). Hence, the edible mushrooms have good nutrition value and could be a good addition to diet of the Ethiopian people.

Keywords: Mushroom, Wild, Cultivated, Proximate, Amino Acid

1. Introduction

Fruiting bodies of mushrooms are appreciated, not only for texture and flavor but also for their chemical and nutritional characteristics (Manzi, Aguzzi, Vivanti, Paci, & Pizzoferrato, 2001). Mushrooms are valuable healthy and nutritious foods, low in calories and high in vegetable proteins, vitamins, iron, zinc, selenium, sodium, chitin, fibers and minerals (Mendil, Uluozlu, Hasdemir, & Caglar, 2004; Ouzouni, Veltsistas, Paleologos, & Riganakos, 2007). Mushrooms have also been reported as therapeutic foods, useful in preventing diseases such as hypertension, hypercholesterolemia, atherosclerosis and cancer. These functional characteristics are mainly due to their chemical composition (Crisan & Sands, 1978; Manzi *et al*., 2001). In general, the fruiting bodies of mushrooms, on dry weight basis, contain about 56.8% carbohydrate, 25% protein, 5.7% fat and 12.5% ash (Demirbas, 2002; Mendil *et al*., 2004). However a number of factors usually influence the nutritional composition of mushrooms. These factors include growing site, type of substrates, mushroom types, developmental stages and part of the fungal samples analyzed (Díez & Alvarez, 2001; Sanmee, Dell, Lumyong, Izumori, & Lumyong, 2003). Although there are a number of reports on the nutritional value of edible mushrooms on literature, there is little or no information available on wild edible mushrooms of Ethiopia, especially collected from southern part like Kaffa and Asosa.

Moreover, since estimation of crude protein is an indirect assay for total amino acids that is affected by the varying levels of non-protein nitrogen present in the sample, the quantitative determination of total amino acids present after acid hydrolysis undoubtedly gives a more accurate evaluation (Weaver, Kroger, & Kneebone, 1977). By far, the predominant methods for determination of amino acids in foods are based on HPLC (Kivi, 2000). However, amino acids are difficult to analyze as-is with sufficient sensitivity and selectivity. In order to increase sensitivity, their analysis normally includes a derivatisation (pre-column or post-

column) step in which the amino acids react with a precursor to yield a strong UV/Visible chromophore or a fluorescent compound (Shimadzu, 2013). Usually, techniques based on ion-exchange separation coupled with post-column derivatization (e.g., with ninhydrin, the "classical" method) are considered more precise (Anders, 2002) than those based on pre-column derivatization and reversed-phase high-performance liquid chromatography (RP-HPLC), because the latter techniques imply extensive sample manipulation before analysis and are affected by the limited stability of the preformed derivatives (Mengerink, Kutlán, Tóth, Csámpai, & Molnár-Perl, 2002). However, such RP-HPLC-based methods have the advantage of being accessible to most analytical laboratories, since they do not require expensive dedicated instruments. In addition, manufacturing of dedicated instruments is being halted. Bartolomeo & Maisano, (2006) recently validated a method of analyzing amino acids by the convenient reversed phase chromatography with pre-column derivatization and uv/vis detection for protein samples.

The aim of the present study was to determine the proximate composition of wild and cultivated edible mushrooms of Ethiopia collected from Kaffa, Asosa and Addis Ababa. Further to measure the quality of the crude protein their amino acid composition was analyzing by using the convenient RP-HPLC-Uv/Vis detection.

2. Materials and Methods

2.1. Description of Sampling Areas and Sites

The three mushroom sampling areas were Addis Ababa, Kaffa zone (site Bonga) and Benishangul gumuz region (site Asosa) of Ethiopia. Addis Ababa is the capital city of Ethiopia and located 9°01 N and 038°45 E. Kaffa zone is situated in the northwestern part of the southern nations, nationalities and people region state (SNNPR) and lies within 07° 00'-7°25'N latitude and 35°55'-36°37'E Longitude. Benishangul gumuz region is located in western parts of Ethiopia located between 09.17° - 12.06° North latitude and 34.10° - 37.04° East longitude.

2.2. Sample Collection and Identification

Identification of the wild edible mushrooms was made by making comparisons with authentic illustrations (Van Der Westhizen & Eicker 1994; Harkonen, Niemela, & Mwasumbi, 2003; Hall, Stefenson, Buchanan, Yun, & Cole, 2003). Moreover, confirmations of the wild mushrooms were made by mycological experts at the department of life sciences at Addis Ababa University.

2.3. Preparation of Samples and Storage

Cultivated mushrooms were dried in oven in the same day they were collected. While the wild mushroom samples were pre-dried on the study areas before transporting to the laboratory using drying rack constructed as illustrated by van der Westhuizen and Eicker (1994). The mushroom samples were cleaned out of forest debris (without washing) with a plastic knife and sliced without separating the cap and the stipe of the mushrooms. Pre-dried samples in the field were further dried in drying oven in the laboratory. The dried samples were milled to fine powder (20 mesh) using smashing machine (FW 100) and kept in plastic bottles until analysis.

2.4. Proximate Analysis

All the analyses were conducted on a dry weight basis.

2.4.1. Moisture Content

Due to the hygroscopic nature of mushroom powders during storage, the moisture content of the dried powder of the mushroom samples were analyzed using a rapid moisture analyzer (Ohaus MB45) heated at 130°C for 15 minutes. Then the moisture correction factor was included in each data analysis so as to block the effect that might come from a difference in the dry matter.

2.4.2. Determination of Crude Protein

The crude protein content of the samples was estimated by the Kjeldhal method (AOAC, 1995), in which the sample was digested with a known quantity of concentrated sulphuric acid in the Kjeltec digestion apparatus. The digested material was distilled after the addition of alkali. The released ammonia was collected in 4% boric acid Kjeltec Automatic Distilling Unit. The resultant boric acid contained the ammonia released from the digested material, and then titrated against 0.1 N HCl, manually. The nitrogen content was determined by multiplying by a factor of 4.38 (Crisan & Sands, 1978) to arrive at the amount of crude protein.

2.4.3. Determination of Crude Fat

The fat content of the samples was determined by Soxhlet, using diethyl ether as a solvent (AOAC, 1995).

2.4.4. Determination of Crude Fiber

Crude fiber content of wild and cultivated edible mushroom samples was determined according to AOAC (2000).

2.4.5. Determination of Crude Ash

The ash content was analysed by weighing the samples before and after burning at 500°C for 24 hours in muffle furnace.

2.4.6. Utilizable Carbohydrate

Utilizable carbohydrates were calculated by difference with the following formula:

Total carbohydrates (g/100g dry weight) =100 - [g protein + g fat + g ash +g fiber]

2.4.7. Energy

Total energy was calculated according to the following equations:

Energy (kcal/100g) =4 x (g protein +g carbohydrate) +9 x (g lipid)

2.4.8. Quality Control

In order to validate the accuracy, reliability and sensitivity of the analytical methods used for protein, fat and ash determination the certified reference material (CRM) BCR-381 (Rye Flour) was used. The CRM was supplied by the Institute for Reference Materials and Measurements (IRMM, Geel, Belgium). The CRM was stored under specified controlled conditions to ensure its stability (Ouzoui, Petridis, Koller, & Riganakos, 2009). Three measurements on the CRM were performed and the results were compared with the certified values. The values for total N, fat and ash in the certified reference material BCR-381 was evaluated using the above experimental methods. The experimental results are in excellent agreement with the certified values.

2.5. Amino Acids Analysis

Protein samples were acid hydrolyzed, then manually derivatized with o-Phthalaldehyde (OPA) and analyzed by RP-HPLC with ultraviolet-visible (UV-Vis) detection, according to a method of (Bartolomeo & Maisano, 2006) with some modifications. Due to the the lack of adequate amount of *Agaricus bispours #1*, *Termitomyces aurantiacus* and *T. microcarpus #2* samples, the amino acid and fatty acid composition of these species were not evaluated.

2.5.1. Acid Hydrolysis

Samples are digested in acid medium to the complete hydrolysis of the protein fraction. Briefly, 100 mg of sample was digested with 3 ml of 6 N HCl at 110°C in heating oven for 24 hours after sealing tubes with nitrogen gas to prevent oxidation. The digested samples were then filtered with Whatman No. 6 and the filtrates were evaporated at 100°C water bath for removing the chlorine gas. Hydrolyzed protein were completely dried with nitrogen gas and re-constituted with 200µl of 0.1 N HCl and kept in the refrigerator till derivatization and HPLC analysis.

2.5.2. Derivatization with OPA

The derivatization was performed manually followed by a cooled (10°C) autosampler. A 60 µL aliquot of the hydrolyzed sample and 60 µl of OPA reagent were taken up with a micropipette and mixed in a HPLC vial. Complete derivatization was achieved by vortexing for 10 seconds. The reaction time after mixing was exactly 2 min. Subsequently, 100 µl of the derivatized sample was transferred into HPLC inserts and 50 µl of the derivatized sample were injected onto reverse phase C-18 column (2.1 mm x 150 mm, 5µm packing column) maintained at temperature of 40°C. The PDA detector where set at λ=338 nm as the preferred wavelength.

2.5.3. HPLC Instrumentation

Amino acid analysis was conducted with Shimadzu HPLC system (Shimadzu, Columbia, MD). The HPLC system consisted of a binary pumping system: pump A (LC-10AD vp) and pump B (LC-10AT vp), a degasser (DGU-14A), an autosampler (SIL-20AC HT), column heater (Brinkmann, CH-30) and UV/Vis diode array detector (SPD-M10A) and system controller (CBM-20A).

2.5.4. Analytical Procedure and Standards

Mobile phase A was 40mM NaH_2PO_4, adjusted to pH 7.8 with 10 N NaOH, while mobile phase B was acetonitrile/methanol/water (45/45/10 v/v/v). The separation was obtained at a flow rate of 2 ml/min with a gradient program that allowed for 2.0 min at 0% B followed by a 16.0-min step that raised eluent B to 53%. Then washing at 100% B and equilibration at 0% B was performed in a total analysis time of 24 min (Bartolomeo & Maisano, 2006).

In order to quantify amino acids present in mushroom samples, the mix standard for protein hydrolysates, AA-S-18 (Fluka) was used. Moreover, individual amino acid standards: Asparagine (Fluka), Alanine (Fluka), Alanine (Sigma), Arginine (Sigma), Aspartic acid (Sigma), Cystine (Calbiochem), Glutamic acid (Sigma), Glutamine (Sigma), Glycine (Sigma), Histidine (Sigma), Isoleucine (Sigma), Leucine (Amersco), Lysine (Sigma), Methionine (Sigma), Phenylalanine (Alfa Sesar), Serine (Sigma), Tyrosine (Sigma), Valine (Alfa Sesar) were used to make the AAS-18 mixture standard list complete and whenever necessary to spike for easy identification of peaks in the mix standard or samples.

2.6. Statistical Analysis

Completely randomized design (CRD) was used. All the experimental results were reported as mean ± standard error (SE) of three parallel measurements. Data were evaluated by using one way variance analysis (ANOVA) and means were separated by Duncan' multiple range test ($p<0.05$) by using SPSS version 15.0.

3. Results and Discussion

3.1. Proximate Composition

When the nutritional value of mushrooms is evaluated, perhaps the most important factor is their moisture content/dry matter, which directly affects the nutrient contents of mushrooms. For example, the moisture of A.bispours varied in one study from 87.2% to 93.5%, a difference of 6.3%. This may seem of little significance; however, the dry matter varied from 6.5% to 12.5%. Hence, the latter will contain 1.97 times as much as nutrients as other (Kurzman, 1997). Thus, in this study to avoid such variability all the samples were oven dried, milled into powder and kept in plastic bottles till analysis. Then the moisture content of each power samples of wild and cultivated edible mushroom was determined using a rapid moisture analyzer so as to block the variability that arises from the nuisance factor. As shown in Table 1 there was very low variation in the moisture/dry matter contents of the mushrooms analyzed in this study (from 8.62 for *T. microcarpus #1* to 13.6 for *T. letestui*). This implies a factor from 1.08 to 1.16 must be multiplied as moisture correction factor for all the other analysis parameters.

Table 1. *Moisture content of the dried wild and cultivated edible mushroom of Ethiopia.*

No	Mushroom Type	Moisture (%)
1	*P .ostreatus*	11.1 ± 0.77
2	*L. eddoes*	9.80 ± 0.68
3	*A. bisporus #1*	10.1 ± 1.32
4	*A. bisporus #2*	12.1 ± 1.20
5	*A. campestris*	8.91 ± 0.63
6	*L. sulphureus*	11.5 ± 0.75
7	*T. clypeatus*	10.9 ± 1.03
8	*T. microcarpus #1*	8.62 ± 0.56
9	*T. aurantiacus*	10.6 ± 0.64
10	*T. microcarpus #2*	10.9 ± 0.72
11	*T. letestui*	13.6 ± 1.1
12	*T. spps*	9.4 ± 0.54

Although fresh, un-dried mushrooms are 85-95% water, they do have significant nutritional value. As fresh mushrooms vary considerably in water content, figures were calculated on a dry-weight basis, which allows a greater consistency of data. To determine an approximate fresh-weight value, results were divide by 10 if the mushroom is very fresh (just picked), by 9 if it has been in the refrigerator for a day or two, or by 8 if purchased from the store (Hobbs, 1986).

Table 2 summarizes the basic chemical composition of the twelve wild and cultivated mushrooms analyzed in this study. The protein content of the samples ranged from 6.84 g/100g (*L. sulphureus*) to 36.7 g/100g (*A. campestris)* on dry weight basis. This implies a person will get 0.68-3.67 g/100g of protein if he/she consumes these mushroom fresh. The protein content of all the samples could have been greater if the factor used to calculate their from nitrogen content was the usual 6.25 than 4.38. Since mushrooms have high proportion of non-protein nitrogen (mainly in chitin), using 4.38 as factor was more appropriate (Crisan & Sands, 1978). Crude protein is thus overestimated in some articles. Mushrooms proved to be good sources of protein compared with green vegetables (Ouzoui *et al*., 2009).

The dry matter of the samples contained fat ranging from 1.34 g/100g (*L. eddoes*) to 5.16 g/100g (*T. microcarpus#1*). This range of fat is in accordance with many literatures which reported that fats in mushrooms usually ranges from 0.2-0.8% of fresh weight, or between 2-8% of the dry weight. According to (Hobbs, 1986) no one will gain weight when dinning on fungi, except when they are sautéed in copious amounts of butter. Crude fat in mushrooms includes representatives of all classes of lipid compounds, including free fatty acids, mono-, di, and triglycerides, sterols, sterol esters and phospholipids (Crisan & Sands, 1978). Of the sterols, various species are especially high in ergosterol, which is the precursor of vitamin D_2 (ergocalciferol) (Mattila, Lampi, Ronkainen, Toivo, & Piironenen, 2002).

There is a big variability in the ash content of the samples (Table 2) ranging from 1.75 g/100g (*L.sulphureus)* to 25.3 g/100g (*T. microcarpus#2).* This difference might be due to the contamination of samples with soil where they are picked up. The gill (spore forming part) of the fruiting body has an opening to trap small soil debris which might not be removed during sample preparation. A similar variation in the ash content of mushroom samples was reported by Vetter (1993). As compared to vegetables, mushrooms proved to be a good source of various minerals as the main constituents in the ash were K and P, whose combined percentage was about 60% (Mattila *et al*., 2001).

Table 2. *Proximate composition (g/100g) of wild and cultivated edible mushrooms of Ethiopia in dwb.*

No	Mushroom Type	Protein (N x 4.38)	Fat	Ash	Fibre	Utilizable Carbohydrate	Energy (kcal/100g)
1	*P .ostreatus*	16.9 ± 0.26f	2.33 ± 0.35d,e	8.26 ± 0.18i	10.2 ± 2.19b,c,d	62.3 ± 0.20c	337.9 ± 9.96c
2	*L. eddoes*	13.4 ± 0.22g	1.34 ± 0.06g	5.66 ± 0.03k	10.9 ± 0.68a,b,c	68.6 ± 0.45b	340.3 ± 3.01c
3	*A. bisporus #1*	25.4 ± 0.54b,c	1.36 ± 0.06g	11.5 ± 0.01f	7.37 ± 0.43d,e	54.4 ± 0.95d	331.4 ± 1.97c
4	*A. bisporus #2*	18.9 ± 0.07e	1.59 ± 0.03f,g	16.9 ± 0.10c	12.3 ± 0.12a,b	50.2 ± 0.07e	290.9 ± 0.20e
5	*A. campestris*	36.7 ± 0.08a	3.45 ± 0.13c	10.3 ± 0.05g	11.9 ± 0.64a,b	37.6 ± 0.45g	328.3 ± 2.90c
6	*L. sulphureus*	6.84 ± 0.13h	2.74 ± 0.05d	1.75 ±0.00l	6.40 ± 0.58e	82.3 ± 0.45a	381.1 ± 2.30a
7	*T. clypeatus*	26.9 ± 0.65b	2.64 ± 0.07d	15.4 ± 0.02d	10.6 ± 0.62a,b,c	44.5 ± 1.20f	309.3 ± 2.86d
8	*T. microcarpus #1*	21.5 ± 1.14d	5.16 ± 0.08a	21.6 ± 0.06b	11.7 ± 0.84a,b,c	40.1 ± 1.71g	292.9 ± 3.92e
9	*T. aurantiacus*	20.5 ± 0.74d,e	2.81 ± 0.03d	6.62 ± 0.03j	8.73 ± 0.64c,d,e	61.3 ± 0.74c	352.6 ± 2.56b
10	*T. microcarpus #2*	24.7 ± 0.97c	4.32 ± 0.31b	25.3 ± 0.12a	13.4 ± 1.31a	32.3 ± 1.39h	266.8 ± 5.49f
11	*T. letestui*	23.6 ± 0.07c	4.12 ± 0.24b	9.75 ± 0.03h	11.1 ± 0.29a,b,c	51.4 ± 0.11d,e	337.1 ± 2.33c
12	*T. spps*	23.9 ± 0.77c	2.01 ± 0.28e,f	15.0 ± 0.05e	10.5 ± 0.79a,b,c	48.6 ± 1.69e	307.9 ± 3.45d

- Means followed by different superscript letters in the same column are are significantly different ($p< 0.05$)
- Data are mean + standard error of three parallel measurements (n=3)
- dwb-dry weight basis

Dietary fiber contents varied (Table 2) from 6.40 g/100g (*L. sulphureus*) to 13.4 g/100g (*T. microcarpus #2).* The dietary fiber in mushrooms is primarily composed of chitin. Chitin is a polymer comprising *N*-acetylglucosamine units and is a constituent of cell walls of most fungi (Miles & Chang, 1997). Chitin is a relatively unusual dietary material, and studies of its dietary effects have been limited (Kurzman, 1997). It may, however, have important physiological properties with respect to human health (Cheung , 1997). Furthermore, some dietary fiber substances, e.g., beta-glucans, have shown powerful antitumor, antimutagenic, and anticancer activity through their stimulatory effects on the immune system (Mattila, Suonpaa, & Piironen, 2000;

Lombardi, 2002).

Utilizable carbohydrate concentrations, calculated by difference varied (Table 2) from 32.3 g/100g (*T.microcarpus# 2)* to 82.3 g/100g (*L.sulphureus)*. Carbohydrates usually account for the prevailing component of fruiting bodies of mushroom (Kalač, 2009). The dry matter of mushrooms contains large amounts of carbohydrate, which, according to the present and previously published data, constitutes the major part of mushroom nutrients. The carbohydrates in mushrooms include polysaccharides such as glucans, mono- and disaccharides, sugar alcohols, glycogen, and chitin (Kurzman, 1997).

The energy value of a food can be estimated based on its crude protein (N x 4.38), fat and carbohydrate using Atwater factors of 4.0, 9.0 and 4.0 kcal/g per component respectively. On the basis of the nutritional analysis, it can be calculated that an edible portion of 100 g of mushrooms on dry weight basis assures, on average, 325 kcal. There is a significant difference ($P < 0.05$) in their energy yield between all species of mushrooms analyzed with highest energy of 381.1 kcal/100g (*L. sulphureus)* and lowest 266.8 kcal/100g *(T. microcarpus #2*) (Table 2). Mushrooms are low-energy foods, only if they are consumed fresh. Otherwise in dry weight these mushrooms could provide high energy.

Overall there is a significant difference ($P<0.05$) in all the proximate compositions of wild and cultivated edible mushrooms of Ethiopia. It seems most of the wild mushrooms have greater nutritional value than the cultivated species. However, *L.sulphureus* has the least of protein, ash and dietary fiber of all the mushroom samples analyzed. There are big differences in the chemical composition of different species of mushrooms. There are differences also within one species depending on the substratum and stage of development (Härkönen *et al*. 2003).

3.2. Amino Acids Analysis

In this study, mushroom samples were acid hydrolyzed with 6 N HCl, then manually derivatized with o-Phthalaldehyde (OPA) and analyzed by RP-HPLC with PDA as detector. Mixtures of OPA-derivatized amino acids can be analyzed with good selectivity and efficiency by reverse-phase HPLC (Jones, 1986). As shown in Figure 2 the amino acids standard mixture (AAS-18-Sigma) were well resolved, although the closely related amino acids such as histidine, glycine and threonine were co-eluted (around 6 minute) and so also valine and methionine (around 9 minute). Thus, the percentage composition for the co-eluted amino acids was reported as a single value in this study. One remedy to avoid the co-elution of amino acids could have been to extend the solvents gradient program at those retention times where co-elution has occurred. However, this would require extending the analysis time and working conditions of the RP-HPLC system. To support this observation, Jones (1986) reported that all of the commonly occurring amino acids can be readily resolved except glycine and threonine. Resolution of these two amino acids is usually a problem, regardless of the column length or particle size of the resin. If baseline resolution of these two amino acids is required, the gradient can be modified, but at the expense of an increased analysis time. Moreover they described other amino acid pairs might also be difficult to resolve in certain OPA-pre column analyzer systems. Thus, this could be seen as one of the limitations of analyzing amino acids with pre-column derivatization with RP HPLC system. However, optimization of a distinct elution gradient conditions such as column length, type of resin, configuration of the the HPLC system, as well as the relative importance of analysis time vs. peak resolution for each individual reverse-phase column could result in better baseline resolution.

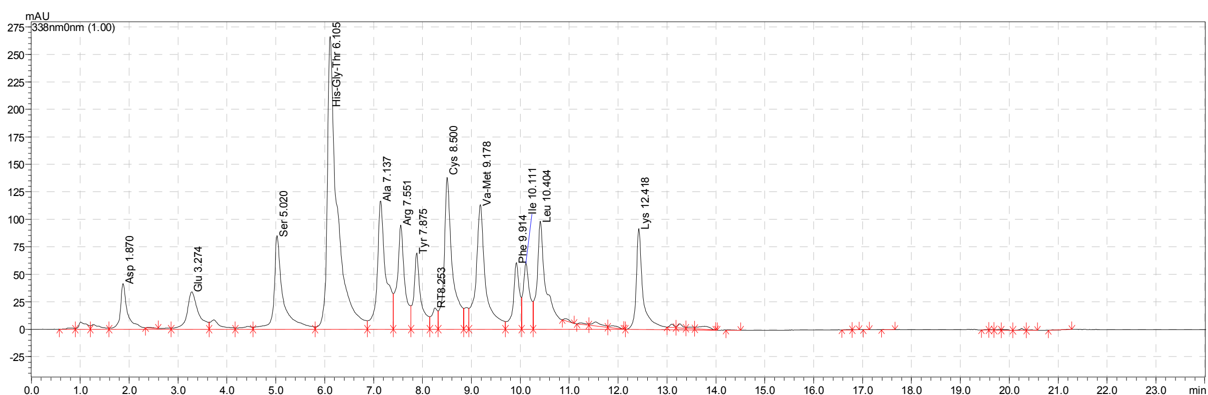

Fig. 1. *Typical chromatogram of the 16 amino acids contained in AAS18 (Fluka) based on RP-HPLC-PDA analysis.*

Identification and quantification of amino acids in the mushroom samples were done by relative comparison of mixture standard against the individual samples. To complete the amino acid list and for verification of peaks, individual standard of all the 18 standards were run and the mixture Perhaps the most appealing aspect of the OPA-precolumn amino acid analysis system is its versatility, since it can be readily adapted to analyze for almost any biological amine from a variety of sources (Jones, 1986).

The percentage composition of the amino acids in the analyzed edible mushrooms species is summarized in Table 3, which reveals that, a total of 18 amino acids was recorded. Tryptophan and proline are the two amino acids not analyzed and reported in this study. The reason for tryptophan is that HCl hydrolysis results in the partial destruction of tryptophan and requires an alternative hydrolysis procedure for accurate

quatitation. For proline, it is because proline is a secondary amine and it does not react directly with the OPA and cannot be determind by this method (Jones, 1986). The amino acids are ranging in percentage (%): Asp (4.87-10.4), Glu (0.64-2.75), Ser (1.46-2.64), Asn (1.90-4.45), Gln (1.06-4.51), His-Gly-Thr (4.05-15.9), Ala (1.07-6.6), Arg (1.62-6.77), Tyr (1.176-10.0), Cys-SS-Cys (1.08-3.67), Val-Met (2.27-6.15), Phe (0.93-1.75), Ile (0.84-1.71), Leu (0.79-9.66) and Lys (0.869-2.37).

Table 3. *Percentage composition of amino acids in wild and cultivated edible mushrooms of Ethiopia.*

No.	Amino acid	Abbreviation	*P.ostreatus*	*L.eddoes*	*A.bispours**	*A.campestris*	*L.sulphureus*	*T.clypeatus*	*T.microcarpus*	*T. letestui*	*Termite spps*
1	Aspartic acid	Asp	10.409	5.930	4.874	6.228	3.304	7.562	8.300	5.948	5.657
2	Glutamic acid	Glu	2.747	1.704	2.980	0.969	0.637	1.417	1.335	1.756	1.751
3	Serine	Ser	2.363	1.463	3.697	2.218	1.878	2.309	2.643	2.118	2.283
4	Asparagine	Asn	4.446	2.667	2.331	1.901	1.900	2.274	2.159	3.920	2.514
5	Glutamine	Gln	1.060	4.506	2.578	1.212	2.459	1.865	1.351	1.526	2.922
6	Histidine	His									
7	Glycine	Gly	9.555	12.297	15.859	4.930	7.076	4.046	9.652	5.351	6.598
8	Threonine	Thr									
9	Alanine	Ala	1.072	4.820	6.606	2.791	1.504	3.203	3.633	1.559	4.891
10	Arginine	Arg	2.114	1.617	5.044	3.718	2.966	3.213	6.769	3.795	1.676
11	Tyrosine	Tyr	1.763	7.059	2.899	3.821	1.773	3.515	2.958	5.142	10.038
12	Cystine	Cys-SS-Cys	1.817	1.076	3.368	2.094	1.704	1.478	3.669	3.516	2.913
13	Valine	Val	4.454	2.267	6.149	4.353	2.378	3.183	3.433	4.704	4.380
14	Methionine	Met									
15	Tryptophan	Trp	Will be lost during acid hydrolysis								
16	Phenylalanine	Phe	1.135	0.927	1.710	1.320	1.450	1.239	1.754	1.396	2.354
17	Isoleucine	Ile	1.321	1.454	1.712	1.220	0.836	0.993	1.074	1.287	1.563
18	Leucine	Leu	1.924	0.790	9.664	1.308	1.032	2.437	2.483	2.533	3.524
19	Lysine	Lys	0.839	0.690	1.512	1.663	1.105	1.344	2.368	2.424	1.082
20	Proline	Pro	2^0 amine and doest react with OPA								

Further, the concentration of each amino acid in g/100g was also calculated by multiplying the percentage contribution of each amino acid with their average crude protein content obtained from the analysis of proximate composition using the Kjeldhal method. Table 4 summarizes the concentration of amino acid of each amino acid in g/100g of dry matter. The 18 amino acids concentration in the mushroom samples ranged: Asp (4.87-10.4), Glu (0.64-2.75), Ser (1.46-2.64), Asn (1.90-4.45), Gln (1.06-4.51), His-Gly-Thr (4.05-15.9), Ala (1.07-6.6), Arg (1.62-6.77), Tyr (1.176-10.0), Cys-SS-Cys (1.08-3.67), Val-Met (2.27-6.15), Phe (0.93-1.75), Ile (0.84-1.71), Leu (0.79-9.66) and Lys (0.869-2.37).

Table 4. *Amino acid profiles of edible mushrooms of Ethiopia (g/100g dry basis) calculated per their crude protein content.*

			P.ostreatus	*L.eddoes*	*A.bispours**	*A.campestris*	*L.sulphureus*	*T.clypeatus*	*T.microcarpus*	*T. letestui*	*Termite spps*
	Crude Protein (g/100g)		**16.9**	**13.4**	**18.9**	**36.7**	**6.84**	**26.9**	**21.5**	**23.6**	**23.9**
No.	**Amino acid**	**Abbreviation**	*P.ostreatus*	*L.eddoes*	*A.bispours**	*A.campestris*	*L.sulphureus*	*T.clypeatus*	*T.microcarpus*	*T. letestui*	*Termite spps*
1	Aspartic acid	Asp	1.76	0.79	0.92	2.29	0.23	2.03	1.78	1.40	1.35
2	Glutamic acid	Glu	0.46	0.23	0.56	0.36	0.04	0.38	0.29	0.41	0.42
3	Serine	Ser	0.40	0.20	0.70	0.81	0.13	0.62	0.57	0.50	0.55
4	Asparagine	Asn	0.75	0.36	0.44	0.70	0.13	0.61	0.46	0.93	0.60
5	Glutamine	Gln	0.18	0.60	0.49	0.44	0.17	0.50	0.29	0.36	0.70
6	Histidine	His									
7	Glycine	Gly	1.61	1.65	3.00	1.81	0.48	1.09	2.08	1.26	1.58
8	Threonine	Thr									
9	Alanine	Ala	0.18	0.65	1.25	1.02	0.10	0.86	0.78	0.37	1.17
10	Arginine	Arg	0.36	0.22	0.95	1.36	0.20	0.86	1.46	0.90	0.40
11	Tyrosine	Tyr	0.30	0.95	0.55	1.40	0.12	0.95	0.64	1.21	2.40
12	Cystine	Cys-SS-Cys	0.31	0.14	0.64	0.77	0.12	0.40	0.79	0.83	0.70
13	Valine	Val	0.75	0.30	1.16	1.60	0.16	0.86	0.74	1.11	1.05
14	Methionine	Met									
15	Tryptophan	Trp	Will be lost during acid hydrolysis								
16	Phenylalanine	Phe	0.19	0.12	0.32	0.48	0.10	0.33	0.38	0.33	0.56
17	Isoleucine	Ile	0.22	0.19	0.32	0.45	0.06	0.27	0.23	0.30	0.37
18	Leucine	Leu	0.33	0.11	1.83	0.48	0.07	0.66	0.53	0.60	0.84
19	Lysine	Lys	0.14	0.09	0.29	0.61	0.08	0.36	0.51	0.57	0.26
20	Proline	Pro	2^0 amine and doest react with OPA								

Mushroom proteins are relatively rich in the amino acids threonine (41–95 mg/g protein DW), valine (36–89 mg/g protein DW), glutamic acid (130–240 mg/g protein DW), aspartic acid (91–120 mg/g protein DW %), and arginine (37–140 mg/g protein DW) but are poor in methionine (1.2–22 mg/g protein DW) and cysteine (16–19 mg/g protein). It has also been reported that lysine, leucine, isoleucine and tryptophan are the limiting amino acids in some edible mushrooms (Cheung 1997; Manzi, Gambelli, Marconi, Vivanti, & Pizzoferrato, 1999; Díez & Alvarez 2001). In general, the differences between the results in this study and those in other reports are assumed to be caused by growth stages, harvesting times, method for conditions of experiments, and so on.

4. Conclusions

The edible mushrooms of Ethiopia were found to be a good source of protein, carbohydrate and essential amino acids. It is also interesting to note that majority of the wild mushrooms (especially *A.campestris* and *Termitomyces*) are consistently more nutritious than their cultivated relatives. However, *L.sulphureus* has less nutritional quality in most of the nutrients than the cultivated ones. In general, wild edible mushrooms of Ethiopia could be a good source of essential nutrients to supplement the diet of the local people.

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