NUTRITIVE VALUE OF SOME WILD EDIBLE MACROFUNGI FROM DEVDAHA, RUPANDEHI DISTRICT, NEPAL

Jha, S. K. and S. Balami

Central Department of Botany, Tribhuvan University, Kirtipur, Kathmandu, Nepal

Abstract

Mature sporophore of common wild edible macrofungi was collected from community forest of Devdaha VDC, Rupandehi, which are commonly harvested by local inhabitants. These macrofungi were then identified and some selected species namely *Termitomyces heimii*, *Lactarius volemus*, *Amanita chepangiana*, *Amanita hemibapha*, *Astraeus hygrometricus* and *Ramaria botrytis* were subjected to proximate nutrient value analysis. Protocol developed by AOAC was thoroughly followed for the proximate estimation of nutrient (protein, lipid, fiber, energy etc.). A total of 24 different wild edible macrofungi was identified from the study sites. Proximate nutrient value analysis showed the protein content of analyzed macrofungi ranges from 8.634 to 48.106%, lipid 1.458-8.315%, fiber 6.451-4.114%, carbohydrate 41.74%-77.346 % and energy content 2335.055-4185.229 Cal/g. The mineral analysis represents presence of good proportion of calcium and phosphorus. Nutritional value of *Lactarius volemus* was found to be higher within the selected macrofungi. Consequently, the wild edible macrofungi show the presence of significant proportion of nutrients and minerals. Thus, these species could address nutritional requirement of local inhabitants and could serve as a potential species for domestication and commercialization.

Key words: Macrofungi, wild edible mushroom, proximate biochemical analysis.

INTRODUCTION

Nepal represents a wide range of ecosystems and habitats because of diverse biogeographic architecture, climatic and altitudinal variations. With this premises, the country also offers a wide array of fungal diversity. The macrofungi are one of them that represent substantial biological resources of the country (Adhikari 2000). Being a key component in nutrient cycling in forest floor, macrofungi also relate to the human livelihood in rural communities, supporting nutritional requirement of natives during their flourishing time (Cunningham and Yang 2012). During the rainy seasons they grow luxuriantly in country's tropical forest to alpine highlands (Adhikari 2006). Varieties of species like *Pleurotus, Agaricus, Ganoderma, Termetes, Ramarai, Lactaria, Amanita, Schizophyllum, Philotus, Psilocybe* etc. are common macrofungi growing (Christensen *et al.* 2008).

Possessing nutrients like proteins, carbohydrates, fats, salts, fibers and vitamins, these macrofungi have potential role in food supplements which has been epitomize by increasing consumption of some commercial mushroom worldwide (Chang 2006). Meantime, wild edible macrofungi have been harvested by ethnic peoples as medicinal and nutritional supplements. Studies suggested that number of wild macrofungi is edible with good organoleptic properties with immense nutraceutical value (Bashir *et al.* 2014, Chang and Chiu 1992). *Ganoderma* sp., *Lentinula* sp. etc. are well known example that have been commercialized successfully. It has been found out that protein content in macrofungi is twice than that of vegetables (Bano *et al*. 1963) and sufficiently higher than that of wheat (Aletor 1995). These macrofungi also possess therapeutic values too. For instance, antimicrobial properties, antioxidants, antitumor, antidiabetic properties, are some of the examples of their therapeutic potential (Chang 2006). Presence of unique metabolic pathway in the formation of varieties of secondary metabolites implied such properties (Zhong and Xiao 2009). Such exquisite dietary value coupled with nutraceutical properties, array of wild mushroom possesses potentiality for usage (Barros *et al.* 2008). But their significance remains still unexplored thoroughly.

The domestication and commercialization of wild macrofungi obviously depend upon their nutraceutical values. Such knowledge can be extracted from the ethnic people living, where they have been using wild macrofungi since time immemorial as medicine and food supplements (Tanti *et al.* 2011, Sarma *et al.* 2010, Adhikari *et al.* 2006). Ethnic knowledge on the consumption on such macrofungi shows that there could be a potential species for domestication and commercialization, diversifying the current mushroom industry.

Thus, this study aims at documenting wild edible macrofungi from one of the ethnic communities of central Terai region of Nepal, and subsequently having nutrient value analysis of some commonly collected wild macrofungi.

MATERIAL AND METHODS

Sample collection and processing

Sporocarps of macrofungi, namely *Termitomyces heimii*, *Lactarius volemus*, *Amanita chepangiana*, *A. hemibapha*, *Astraeus hygrometricus* and *Ramaria botrytis* were collected from the community forest of Devdaha VDC, Rupandehi, Nepal. The sites represent tropical hardwood forest with dominant vegetation of *Shorea robusta*, *Semicarpus anacardium*, *Adina cordifolia* etc. Collection was done between July 2014 and September 2015. The samples were first oven dried at 80ºC for 48 h and pulverized in a grinder. The fine powdered samples were stored into a hermitic flask below 10ºC.

Proximate estimation of nutrients

Standard protocol proposed by AOAC (2000) was followed for determining dry matter content, ash content, crude fiber, protein, carbohydrate, lipid, and mineral contents.

(i) *Dry matter content:* Known weight of samples were heated at 100ºC, then cooled in desiccators and reweight until the difference in weight between the successive weighing was one milligram.

$$\text{Dry matter (\%)} = \frac{\text{Dry sample weight}}{\text{Sample weight}} \times 100$$

(ii) *Ash content:* one g of the oven dried sample was heated to 100ºC for 24 h, reweight, then incinerated in muffle at 550^{o} C for 4-5 h to free carbonaceous material. Again after, cooling followed by drying on asbestos sheet and then in desecrator, sample was reweight until the difference between two successive weighing was less than 1 mg.

$$\text{Total ash content (\%)} = \frac{\text{Weight of ash}}{\text{Weight of sample}} \times 100$$

(iii) *Crude fiber content:* one g of dried sample was boiled with 1.25 % H_2SO_4 for 30 min, filtered, and then washed with DW to neutralize. And then washed with 15 ml of C_2H_5OH (EtOH). The crucible was dried at 110^{o} C to constant weight.

$$\text{Crude fiber (\%)} = \frac{(\text{Sample wt} - \text{Ash wt})}{\text{Sample wt/ DM\%}} \times 100\%$$

(iv) *Crude protein* (Micro – Kjeldahl's method): 0.5 g of sample was treated with conc. H_2SO_4 in a digestion tube with 0.2 g of $CuSO_4$, 0.1 g of K_2SO_4 until it turns green. 10 ml of the sample was then transferred to a steam chamber and 40% NaOH (10ml) was added for digestion. Immediately, the stop-clock was closed and passed through the steam chamber. Then the receiving flask was removed from the condenser outlet and rinsed with water then titrated with 0.028 N HCl till it turns pink.

$$\text{Crude protein (\%)} = \frac{14 \text{ x Normality} \times (\text{Reading point} - \text{Blank})}{\text{Sample wt} \times \text{DM\%}} \times 6.25 \times 1000000$$

(v) *Lipid content*: Fat was extracted with petroleum ether in Shoxlets apparatus. After extraction, thimbles was remove, air-dried overnight and then dried at 70^{o}C for 24 h, cooled and dried in desiccators and reweighed.

$$Crude\ fat\ (\%) = \frac{(\text{Flask weight} + \text{Fat weight})_{\text{Flask weight}}}{(\text{Sample weight x Dry matter }\%)} \times 10000$$

(vi) *Energy content*: A weighed sample was placed in a heavy steel container (bomb). After bomb was charged with oxygen, sample was ignited. The change in the temperature of the water was observed.

$$\text{Energy (cal/g)} = \frac{\text{Temp. change} \times (2000 \times \text{standard})}{\text{Sample wt/DM}\%} \times 1000000$$

(vii) *Carbohydrate content*: Total carbohydrate amount was calculated by employing the following formula.

Carbohydrate = 100 - (Moisture + Protein + Fat + Ash + Fiber) g.

(viii) *Mineral content:* Oven dried sample was incinerated in a muffle furnace (550°C) for 4-5 h to free from carbonaceous material, then cooled. 10 ml HCL (36%) and 30ml DW were added to the aliquots then heated for ½ h and cooled to RT and filtered. Filtrates were then diluted to 100 ml. For calcium content, 10 ml aliquot ash solution was taken, three drops of 0.02% methyl red, five ml of 3% ammonium oxalate and 2-4 g urea were added. Solution was heated until colour was changed and left for overnight and filtered with a piece of filter paper and washed by ammonium solution (7-8 times). Then, the sample with filter paper was taken in a same beaker and after adding hot (about 60° C -80°C) 50 ml H_2SO_4 (1:24) the solution was added. Finally the solution was titrated with 0.004N $KMnO_4$ solution. Percentage of calcium was calculated as:

$$Calcium\ content\ (\%) = \frac{(\text{Reading point} - \text{Blank}) \times (0.1002\text{xNormality}) \times \text{Dilution factor}}{\text{Sample wt/DM}\%} \times 100000000$$

For phosphorus content, l0 ml aliquot was taken and added to 10ml of NH_4VO_3, 10ml of nitric acid (1:3) and 10 ml of $(NH_4)6Mo7O_{24}.4H_2$ O and diluted to 100 ml then allowed to stand for 20 to 30 minutes. The absorbance at 430 nm wavelength was measured. The phosphorus content was calculated as:

$$\text{Percentage of Ph}os\text{phorus} = \frac{\text{Content X Dilution}}{\text{Sample wt X100/Dm}\%} \times 100, \text{ Where, } content = \frac{\text{Absorbance} - \text{X+Value}}{Y - value} \times 100.$$

Statistical analysis

The data were analyzed with Microsoft excel 2007, where the mean value of the percentage data were calculated. Further rank analysis was made on the various nutrient content (6 for the highest values and 1 for the lowest value), and total rank bar graph was plotted.

RESULTS AND DISCUSSION

Frequent visit to the sites and questionnaire with local inhabitants – especially key informants, list of wild edible macrofungi were obtained (Table 1). Further Melzer's test was used to have tentative information on their edibility status. Selected six macrofungi were then subjected for proximate nutrient analysis. Altogether 24 different macrofungi were identified as listed in the Table 1, which were found to be commonly harvested by the locals for food supplements and medicinal purpose.

Varieties of macrofungi were reported as edible in harmonious with many other such ethnomycological surveys and reporting in world and Nepal (Boa 2004, Christensen *et al.* 2008). The sites harbor common edible mushrooms like *Pleurotus oestratus, Agricus* sp, *Volverialla volvecea* which have been industrialized commercially. Additionally, medicinal mushrooms like *Ganoderma lucidulum* are also recorded. The locals responded that the macrofungi *Termitomyces hemii* possess good organoleptic properties among the edible collection during flourishing time. Survey at the field also suggests that harvesters have some kind of knowledge on the sustainable harvest of these macrofungi, for instance not collecting all the mature caps.

Table 1. List of wild edible macrofungi from the study sites with their local name and family.

Mushroom species	Local name	Family
Agaricus sp.	Gobre chayau	Agaricaceae
Amanita hemibapha	Salle chyau	Amanitaceae
Astreatus hygrometricus	Kathe chyau	Astraeaceae
Auricularia auricular	Bilarickan	Auriculariaceae
Boletus sp.	--	Boletaceae
Cantharellus sp.	Chamre/m ude chyau	Cantherallecae
Cantharellus sp.	--	Cantheralleceae
Clitocybe geotropa	Kathchiwa	Tricholomataceae
Fomitopsis pinicula	Kathchiwa	Polyporaceae
Ganoderm lucidum	Kathe chayau	Ganodermataceae
Lactaria volemnus	--	Lactariaceae
Pleurotus oestratus	Kathchiwa	Pleurotaceae
Pleurotus conucopiae	--	Pleurotaceae
Polyporus durus	Kathey chayau	Polyporaceae
Polyporus sp.	--	Polyporaceae
Pycnoporus cinnabarinus	Kane chyau	Polyporaceae
Ramaria botrytis	--	Clavariaceae
Russula emitica	Kali chyau	Russulaceae
Russula delica	--	Russulaceae
Schizophyllum commune	Pankha chayau	Schizophyllaceae
Scleroderma bovista	Dalle chyau	Sclerodermataceae
Scleroderma sp.	Gangadhur	Sclerodermataceae
Termitomyces hemii	Bhabnethi/bagale chyau	Tricholomataceae
Volverialla volvecea	Purchhatta	Plutaceae

Based on AOAC (2000) protocol, various parameters signifying nutrient value of selected macrofungi showed different proportion of nutrient contents as shown in Table 2.

Table 2. Nutrient contents of selected macrofungi.

Parameters(%)	Macrofungi species					
	A. chepangiana	*A. hemibapha*	*A. hygrometricus*	*L. volemus*	*T. heimii*	*Ramaria botrytis*
Dry matter	93.88	94.701	96.911	95.185	93.443	92.416
Ash	0.796	1.001	0.357	0.55	0.350	0.377
Crude fibre	10.083	12.005	6.451	22.772	21.885	9.562
Crude protein	25.933	27.947	16.219	8.634	48.106	29.5148
Crude fat	8.314	8.043	2.742	5.833	1.373	1.458
Carbohydrates	55.753	55.711	76.128	77.346	41.74	59.491
Energy (cal/g)	3796.478	2335.054	3740.706	4000.231	3987.053	4185.229
Calcium	0.853	0.761	0.785	1.389	0.4289	0.520
Phosphorus	0.632	0.626	0.251	0.343	0.641	0.477

Among the analyzed mushroom sample, dry matter content was on relatively higher in *A. hygromatricus* (96.91%) and least in *R. botrytis* (92.416%). The dry matter content of macrofungi growing in forest floor is the function of the habitat variability – the moisture content in the soil, the salt concentration in soil etc. Dry matter (DM) content analysis on button mushroom suggests that with increasing osmolarity in soil the dry matter content increased (Van Loon *et al.* 2000). However, increase dry matter content in macrofungi also correlates with the lower density of fruiting bodies in culture. But, the firmness of the fruiting bodies remains relatively higher suggesting the good quality of the

mushroom. As such, DM analysis results suggest all the tested macrofungi could possess minimal postharvest loss.

Percentage of ash content on DM basis of the tested macrofungi varied from 0.796% in *Amanita chepangiana* to least of 0.35% in *T. heimii*. Primarily macrofungi growing on wood shows higher values of ash content (Hoa *et al.* 2015), consequently macrofungi growing in recalcitrant substrate possess high mineral content/ ash content. As such, the dietary value of *A. chempangiana* is relatively higher as compared to other tested species as percentage of ash content shows higher mineral content. Meantime, the higher ash content has been correlated with the decrease in amino acid contents in meat but data don't represent such instance in the tested macrofungi (Table 2).

The crude fiber is a group of indigestible carbohydrate that possesses significant dietary value (Wang *et al.* 2014). In the present study, the crude fiber content of different mushroom sample had found in range of 6.451 to 22.772%. The crude fiber percentage in *L. volemus* had the highest percentage of crude fiber while *A. hygrometricus* (6.451%) had the lowest value. The above result showed appreciable amount of crude fiber present in different wild edible mushroom which is good for human consumption. Thus, *L. volemus*, followed by *T. hemii* possess good dietary values.

The crude protein (CP) content of the different wild mushrooms has varied greatly on species to species (Table 1). The dry weight percentage of CP content was the highest in the *T. hemii* (48.106%) followed by *R. botrytis* (29.514%), *A. hemibapha* (27.947%), *A. chepangiana* (25.933%), *A. hygrometricus* (16.219%) and *L. volemus* (8.634%). The crude protein content of edible mushrooms is usually high, but varies greatly and is affected by various factors such as species and stage of development, habitat and environmental conditions (Ayaz *et al.* 2011) as well as methods of drying technique also alter the nutritional composition of mushrooms species (Adedayo *et al.* 2010). The finding of this study agrees with the study that crude protein content of *R. flava* (28.32%) and *A. hemibapha* (25.87%) (Giri and Rana 2009). Thus these wild edible macrofungi are the good source of alternative protein supplement to the rural people due to its high protein content.

Macrofungi belonging to Basidiomycota generally show marked differences in lipid content (Miric *et al.* 1985). The total crude fat content of analyzed sample ranges between 0.225% and 8.044%. *Amanita chepangiana* (8.314%) had highest value of crude fat followed by *Amanita hemibapha* (80.043%) and least in *T. heimii* (0.225%). The variation of crude fat content of the different wild edible mushrooms agrees with the previously reported data on sporocarps of higher basidiomycetes (Chang and Miles 2004). The results were comparable with previous analysis too (Jha and Tripathi 2012, Giri and Rana 2009). As such consumption of these mushroom were good for human health due to low crude fat content.

Carbohydrate content of selected macrofungi samples showed the presence of highest carbohydrate content in *L. volemus* (77.346%) and lowest in *T. heimii* (41.74%). The data is coupled with energy content in these macrofungi, where energy content is also highest in *L. volemus* (4000.231 Cal/g). Usually, energy content of the mushroom generally low, which allowed them to be used in low energy diets.

Studies suggests wild macrofungi as good source of minerals, containing macro- elements like calcium, magnesium, potassium, sodium and phosphorus (Agrahar-Murugkar and Subbulakshmi 2005). Consequently calcium and phosphorus content in tested fungi represents similar instance. The calcium content (%DM) ranged from 0.626% in *T. hemii* to 1.389% in *L volumus*. Phosphorus (%DM) level was the highest in *T. heimii* (0.641%) followed by *A. chepangiana* (0.632%) and so on (Table 2) The range of mineral content of present findings was similar to the previous findings (Pandey and Budhathoki 2007).

L. volemus possess highest rank of all the added components of nutrients, suggesting it as a potential macrofungi for domestication and cultivation. Although organoleptic properties limit its preference, the dietary value was relatively good.

Different wild edible macrofungi showed considerable proportion of nutrient content. They possess a rich amount of protein and low fat content. However there are low ash and fiber content than that of animal and plant origin. Among the tested species, *L. volemus* contain relatively higher proportion of nutrient content, so this species could have potential for domestication and mass cultivation. In overall, rich proximate nutritional composition that makes these wild macrofungi as a choice food for local people of study area. So wild edible mushroom are the very important food that may fulfills required amount of nutrient (carbohydrate, protein, energy, vitamins and minerals) supplement for the rural people. Hence, these kinds of information should help to conserve the plant resource and also help local people to know more about the nutritional and medicinal value and uplift people's livelihood.

ACKNOWLEDGMENTS

This work was supported by the National Academy of Science and Technology, Khumaltar, Lalitpur, Nepal. We would like to acknowledge Prof. Dr. Promod K. Jha (former head of the Central Department of Botany, Tribhuvan University, Kirtipur, Kathmandu, Nepal), and also thankful to Ghanashyam Chaudhary and Uttam Osti.

REFERENCES

Adedayo, M. R., G. Olasehinde and A. A. Ajayi. 2010. Nutritional value of some edible mushrooms from Egbe farmland, West Yagba Local Government Area, Kogi State, Nigeria. *Afr. J. Food Sci.* **4**(5): 297-299.

Adhikari, M. K. 2000. *Mushrooms of Nepal*. PU Printers, Batisputali, Kathmandu, Nepal. 236 pp.

Adhikari, M. K., S. Devkota and R. D. Tiwari. 2006. Ethnomycolgical knowledge on uses of wild mushrooms in Western and Central Nepal. *Our Nature.* **3**(1): 13-19.

Agrahar-Murugkar, D. and G. Subbulakshmi. 2005. Nutritional value of edible wild mushrooms collected from the Khasi hills of Meghalaya. *Food Chem.* **89**(4): 599-603.

Aletor, V. A. 1995. Compositional studies on edible tropical species of mushrooms. *Food Chem*. **54**(3): 265-268.

AOAC. 2000. *Association of Official Analytical Chemists*. Official methods of analysis, Washington DC. USA.

Ayaz, F. A., H. Torun, A. Ozel, M. Col, C. Duran, E. Sesli and A. Colak. 2011. Nutritional value of some wild edible mushrooms from the Black Sea region (Turkey). *Turk. J. Biochem*. **36**(3): 213-221.

Bano, Z., S. Srinivasan and H. Srivastava. 1963. Amino acid composition of the protein from a mushroom (*Pleurotus* sp.). *Appl. Microbiol. J.* **11**(3): 184-187.

Barros, L., T. Cruz, P. Baptista, L. M. Estevinho and I. C. Ferreira. 2008. Wild and commercial mushrooms as source of nutrients and nutraceuticals. *Food and Chem. Toxicol*. **46**(8): 2742-2747.

Bashir, A., N. Vaida and M. D. Ahmad. 2014. Medicinal importance of mushrooms: A review. *Int. J. Adv. Res.* **2:** 1-4.

Boa, E. 2004. *Wild edible fungi: A global overview of their use and importance to people*. Food and Agriculture Organization (FAO), Rome, Italy., pp. 1-156.

Chang, S. T. 2006. The World Mushroom Industry: Trends and Technological Development. *Int. J. Med. Mushr*. **8**(4): 297-314.

Chang, S. T. and P. G. Miles. 2004. The nutritional attributes of edible mushrooms. In: S. T. Chang and P. G. Miles (eds.). *Mushrooms: Cultivation, Nutritional Value, Medicinal Effect, and Environmental Impact.* 2nd ed. CRC Press, Inc. Boca Raton, Florida, USA., pp. 27–37.

Chang, S. T. and S. W. Chiu. 1992. Mushroom production-An economic measure in maintenance of food security. In: E. J. DaSilva, C. Ratledge and A. Sasson (eds.). *Biotechnology: Economic and Social Aspects*. Cambridge University Press, Cambridge, UK., pp. 110-141.

Christensen, M., S. Bhattarai, S. Devkota and H. O. Larsen. 2008. Collection and use of wild edible fungi in Nepal. *Econ. Bot*. **62**(1): 12-23.

Cunningham, A. B. and X. Yang. 2012. *Mushrooms in Forests and Woodlands: Resource Management, Values and Local Livelihoods*. Taylor and Francis, Routledge. 240 pp.

Giri, A. and R. Rana. 2009. Ethnomycological knowledge and nutritional analysis of some wild edible mushrooms of Sagarmatha National Park (SNP), Nepal. *J. Nat. Hist. Mus*. **23**: 65-77.

Hoa, H. T., C. L. Wang and C. H. Wang. 2015. The Effects of Different Substrates on the Growth, Yield, and Nutritional Composition of Two Oyster Mushrooms (*Pleurotus ostreatus* and *Pleurotus cystidiosus*). *Mycobiology*. **43**(4): 423-434.

Jha, S. K. and N. N. Tripathi. 2012. Comparative nutritional potential of three dominant edible and medicinal macrofungi of Kathmandu valley, Nepal. *Am. J. PharmTech Res*. **2**(3): 1036-1042.

Miric, O., V. Z. Lalic and D. I. Miletic. 1985. The composition of some lipid fractions (phospholipids, triglycerides, free fatty acids, sterols) of wild growing edible mushrooms. *Hrana i ishrana.* **26**: 5-8.

Pandey, N. and U. Budhathoki. 2007. Protein determination through Bradford's method of Nepalese mushroom. *Sci. World.* **5**(5): 85-88.

Sarma, T. C., L. Sarma and B. N. Patiri. 2010. Wild edible mushrooms used by some ethnic tribes of western Assam. *The Bioscan*. **3**: 613-625.

Tanti, B., L. Gurung and G. C. Sarma. 2011. Wild edible fungal resources used by ethnic tribes of Nagaland, India. *Indian J. Tradit. Know*. **10**(3): 512-515.

Van Loon, P. C. C., H. A. T. I. Swinkels and L. J. L. D. Van Griensven. 2000. Dry matter content in mushrooms (*Agaricus bisporus*) as an indicator of mushroom quality. *Mushroom Sci.* **15**: 507-513.

Wang, X. M., J. Zhang, L. H. Wu, Y. L. Zhao, T. Li, J. Q. Li and H. G. Liu. 2014. A mini-review of chemical composition and nutritional value of edible wild-grown mushroom from China. *Food Chem*. **151**: 279-285.

Zhong, J. J. and J. H. Xiao 2009. Secondary metabolites from higher fungi: discovery, bioactivity, and bioproduction. In: J. J. Zhong, F. Wu Bai and W. Zhang (eds.). *Biotechnology in China*. Springer Heidelberg, Berlin, Germany., pp. 79-150.