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Analysis of antioxidant activities of common wild edible mushrooms employing cuprous assays: A comparative study

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Abstract

Ranchi is the state capital of Jharkhand. Ranchi district nurtures myriads of micro climatic conditions and habitats which provide suitable environment for the growth of different types of wild edible mushrooms especially during monsoon.These mushrooms are widely distributed in this district and it is an important source of nutrient, health, and income generation. The current study was pivoted at the antioxidant activities of some of the mushrooms. A total of 7 most commonly consumed wild edible mushrooms were analyzed using the cuprous assay. The data from the cuprous assay showed that all studied wild edible mushrooms possess antioxidant activities. Among all the edible species, the dried ethanolic extracts of *Termitomyces heimii* Natrajan showed highest antioxidant content (120.6 mg/ml) followed by Astraeus *asiaticusWatling* (115.3 mg/ml,)*Pleurotus ostreatus* P. Kumm(90 mg /ml), *Termitomyces clypeatus R, Heim* (87.6 mg/ml), *Boletus edulis* Fr. (78.8 mg/ml),*Termitomyces microcarpus* Berk & Broome (76.6 mg/ml) and *Volvariella volvacea* Speg. (70 mg/ml). Thus, wild edible mushrooms may have prospective as natural antioxidants.

Keywords: Wild edible mushrooms; Health; Antioxidant activities

1 Introduction

When rain showers then the mushrooms around us come alive. The term "wild" mention to mushrooms that have not been cultivated and have grown naturally. Wild mushrooms are seasonal fungi, which occupy diverse niches in nature in the forest ecosystem. They are the indicators of the forest life support system. Mushrooms are used as food for nutritive value and medicinal values as dietary supplements which produce high quality and economic value to the world market of mushrooms. Wild mushrooms have been used not only for cuisine but also been used for the treatment of several diseases. Mushrooms that are considered to have some medicinal applications are considered medicinal mushrooms such as, *Ganoderma lucidum, Cordyceps sinensisetc*. Wild medicinal fungi are also collected and used in most countries.The momentous therapeutic and physiological properties of mushrooms are immune system improvement, maintenance of equilibrium and regulation of biorhythm, cure & prevention of various diseases, and improvement from life-threatening diseases such as tumors, stroke, and heart diseases[1]. In contemporary terminology, mushrooms are also called therapeutic foods or nutraceuticals due to their pharmacological properties[2,3]. The work of many researchers shows the anti-microbial, anti-viral, and anti-tumor properties of mushrooms[4,5,6] . Their hypocholesterolemia and anti-inflammatory properties were also demonstrated[7]. Edible mushrooms are a rich source of bioactive compounds due to their antioxidant activities [8,9]. These days, various species of mushrooms used in thousand-year-old traditional medicine practices are under intense study by ethnobotanists and medical researchers[10]. Maitake, Shiitake, Chaga, and reishi are prominent among those being researched for their potential anti-cancer, anti-viral, or immunity-enhancing properties and various other medicinal effects. The content and type of biologically active substances may vary considerably in edible mushrooms; their concentrations of these substances are

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affected by differences in strain, substrate, cultivation, developmental stage, age, storage conditions, processing, and cooking practices [11]. In addition to the nutrient values found in edible mushrooms, some have been found to comprise important amounts of bioactive compounds [12].

Keeping this fact in mind, the present work has been chosen and giving emphasis on the antioxidant activities and medicinal properties of some wild edible mushrooms which can safely be used as Nutraceutical.

2 Material and methods

The total antioxidant activities of wild mushrooms were analyzed by the CUPRAC Method (ozyurek et.al; 2011) [13]. The cupric reducing antioxidant capacity (CUPRAC Method) is based on the absorbance measurement of CU (1) – Neocuproine chelate formation as a consequence of the redox reaction of chain-breaking antioxidants with the CUPRAC reagent, which provides a color that is measured at 450 nm, Neocuproine gives yellow colour and intensity of colour depends on the amount of CU (II) that is reduced to CU (I). Thus the degree of change of colour either decreases or increases absorbance at 450 nm is related to the concentration of antioxidants in the sample.

2.1 Preparation of mushroom extract

Dried mushroom powder (5 gm of sample) was soaked with 50 ml of ethanol. The beaker was covered with foil to minimize the rate of evaporation and kept for 48 hrs. After 48 hrs solution was filtered by using filter paper & filtrate was collected in a beaker it then was covered with muslin cloth and kept in the room for evaporation of the solvent. After evaporation, the remaining extract was used for antioxidant assay and the remaining extract was then stored at 4 degrees celsius for further use.

2.2 Chemical used

Copper Chloride ($CuCl_2$) Neocuproine ($C_{14}H_{12}N_2$)
Ammonium Acetate ($NH_4CH_3CO_2$)

2.3 Procedure for antioxidant assay by CUPRAC Method

Briefly, about 1 ml each of prepared Copper Chloride, freshly prepared Neocuproine, and Ammonium acetate were added to the test tube, then antioxidant sample solution and water were added to the test tube (in which 1 ml each of Copper Chloride + Neocuproine + ammonium acetate was added) to make the final volume up to 4.75ml. Test tubes with all the mixtures were kept in dark for 30 minutes and O.D was taken against a reagent blank at 450 nm (blank is prepared without putting extract or ascorbic acid as standard).In blank 1 ml each CUCl2, Ammonium Acetate&Neocuproine were added. The linear calibration curves of the tested antioxidants as CUPRAC absorbance versus concentration were drawn.

2.4 Antioxidant activities of different wild edible mushrooms

Average O.D of standard compound ascorbic acids at 450 nm and its calibration standard curve are represented in the Table1 and Figure 1 respectively.

Table 1 Absorbance of standard compound, ascorbic acids at 450 nm wavelength

Conc.(µg/ml)	Absorbance (Mean) λmax =450 nm
10	0.112
40	0.228
80	0.372
120	0.497
160	0.551
200	0.71

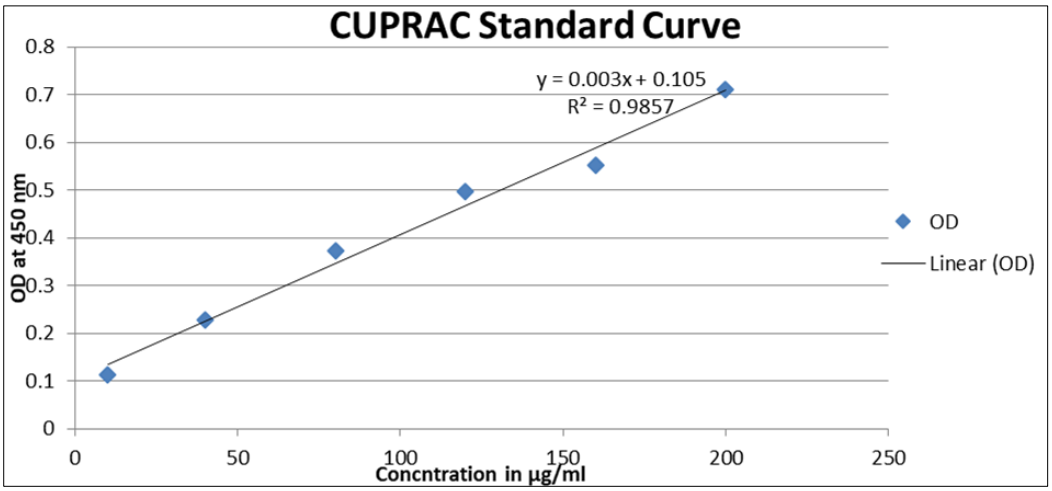

Figure 1 Calibration standard curve of ascorbic acids for determination of antioxidant activities in wild edible mushroom

3 Results and discussion

Wild edible mushrooms are one of the major natural deposit on which the local people of Ranchi district depends and they play a vital role in providing nourishment. Being latent qualities of wild mushrooms with antioxidant activities have not been studied much, the present study was therefore aimed at determining the antioxidant activities of some common wild species of mushrooms. The results of antioxidant activities analysis of wild edible mushrooms are presented in figure 2 & 3 and table 2.

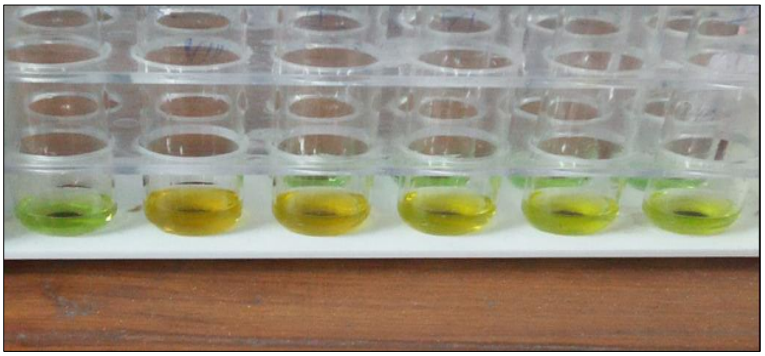

Figure 2 Antioxidant analysis

Table 2 Antioxidant activities of wild mushrooms in (mg/ml)

Concentration of sample mg/ml	Antioxidant activities of wild mushrooms in (mg/ml)						
	T. heimmi	***T. clypeatus***	***T. microcarpus***	***Astraeus asiaticus***	***Volvariella volvacea***	***Boletus edulis***	***Pleurotus ostreatus***
40	37.6	32.3	30	36.6	25.3	32	36.6
80	71.6	58.6	40	69	35.6	44.6	63.3
120	89.3	68.6	63.3	86.6	58	65.3	82
180	120.6	87.6	76.6	115.3	70	78.8	90
200	156.3	110	92..6	148.3	90.6	96.6	106.6

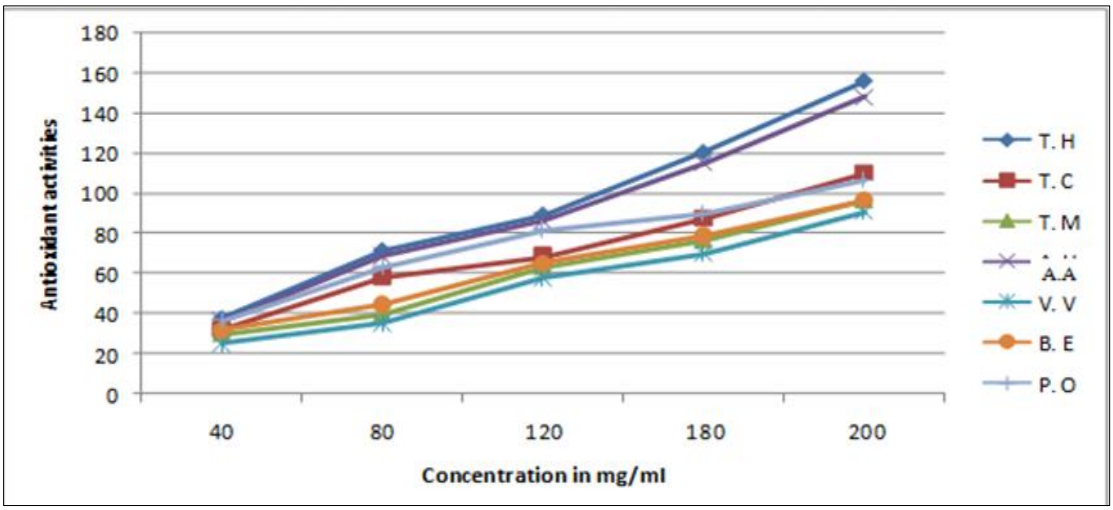

Figure 3 Antioxidant activities present in the seven spp. of wild edible mushrooms in mg /ml of dry weight

Figure 4 Dried Mushroom Powder soaked with ethanol

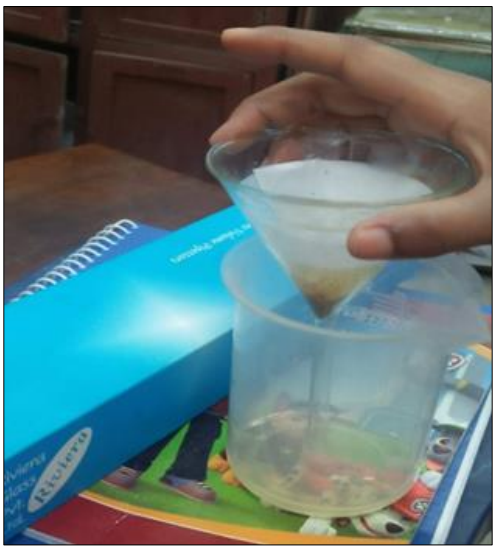

Figure 5 Collection of filtrate

Dried powdered collected wild edible mushrooms were ethanolic extracted and examined (Fig.-4,5,6). All mushrooms studied contained antioxidant activities. Among all the edible species , the dried ethanolic extracts of *Termitomyces heimii* showed highest antioxidant content (120.6 mg/ml) followed by Astraeus asiaticus (115.3 mg/ml,) *Pleurotus ostreatus* (90 mg/ml), *Termitomyces clypeatus* (87.6 mg/ml) ,*Boletus edulis* (78.8 mg/ml), *Termitomyces microcarpus* (76.6 mg/ml) and *Volvariella volvacea* (70 mg/ml). Results from cuprous assay showed that all studied wild edible mushrooms possess antioxidant activities. So, wild edible mushrooms may have prospective as natural antioxidants.

Figure 6 Mushroom Extract

Explication of the organism to free radicals has led to the development of autogenous defence techniques to eliminate them. These defences were the response of evolution to the inevitability of ROS production in aerobic conditions. Natural products with antioxidant activity may help the autogenous defence technique. In this perspective, the antioxidants present in the diet suppose of vital importance as possible protector agents reducing oxidative damage. Particularly, the antioxidant properties of wild mushrooms have been extensively studied by the research group and many antioxidant compounds extracted from these sources have been identified, such as phenolic compounds, tocopherols, ascorbic acid, and carotenoids [14] .The work is per the observation made by some researchers [15,16,17].

4 Conclusion

Comparative antioxidant activities of wild edible mushrooms were found to vary from 70 mg/ml to 120.6 mg/ml, *Volvariella volvacea* (70 mg/ml) was found to show the lowest antioxidant activities while the highest antioxidant activities were shown in *Termitomyces heimii* (120.6 mg/ml). Thus results from the cuprous assay showed that all studied wild edible mushrooms possess antioxidant activities. So, they might be used precisely in diet and advocate health, taking advantage of the additive and synergistic effects of all the bioactive compounds present.

Compliance with ethical standards

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Disclosure of conflict of interest

The authors declare no conflict of interest.

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