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CHARACTERIZATION AND UTILIZATION POTENTIALS OF MUSHROOM SPECIES GROWN ON WASTE CARDBOARD COMPOST

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ABSTRACT

This study assessed the quality of mushroom species that grown naturally on the waste cardboard compost at the stage of maturity. Since they were not intentionally cultivated, their characterization became necessary. Physical observation, laboratory analysis and literature search were used to classify the mushrooms. The fruiting bodies of the mushrooms were analyzed in a laboratory for their macro mineral and nutritional contents, using standard method (APHA). At maturity, two species of mushrooms that colonized the compost were *Inocybe margaritispora* and *Corprinus narcoticus*. Results of chemical analysis indicated organic carbon (35.35 ± 0.04% and 33.28 ± 0.00%); nitrogen (5.58 ± 0.02% and 33.28 ± 0.00%) and phosphorus (0.33 ± 0.02% and 0.24 ± 0.01%) for the two species respectively. With the exception of potassium, all the parameters were found to be higher in *Inocybe margaritispora*. Lead and cadmium were (18.35 ± 0.05 mg/kg and 31.30 ± 0.15 mg/kg) and (1.88 ± 0.03 mg/kg and 1.37 ± 0.04 mg/kg) respectively, which exceeded the stipulated international standards. Due to the high levels of heavy metals and lower macro nutrient contents, it is therefore not advisable to utilize them for human consumption. They can, however, find use in some other biotechnology–based functions.

INTRODUCTION

Mushroom cultivation from wastes is another way, second to compost production, by which waste managers contribute to the food security of a nation. Mushrooms are very rich in protein and also compose of sugars, myco-cellulose, fats, carbohydrates, all essential minerals (Jiskani, 2001) and vitamins such as D, C, B1, B5, B6, niacin, biotin thiamine and riboflavin (Chang and Buswell, 1996; Miles and Chang, 1997; Buigut, 2002). Proper identification of the mushroom is, however, extremely important. Unfortunately, many mushrooms are difficult to identify even by a trained mycologist and a biologist trained in the study of mushrooms (H.O.R.S.E. Inc, 2010). Massive liver and kidney failure can cause human death in a matter of minutes if poisonous mushrooms are consumed. Death from other species, even so-called "safe" mushrooms, has occurred in very young children or in very ill adults (David et al., 2012). Prior to the commercial cultivation of mushrooms in Nigeria, a large proportion of the people relied on collection of wide mushrooms with the occasional dare consequences of mortality (and less frequently morbidity) as a result of consumption of poisonous species of mushrooms (Uraih and Izuagbe, 1990).

Earlier on, many studies have been carried out on mushroom in Nigeria and worldwide. Ogundana and Fagade (1981) indicated that mushroom was about 16.5% dry matter out of which 7.4% was crude fiber, 14.6% was crude protein and 4.48% was fat and oil. Results of proximate analysis of four edible species of mushroom collected from Akoko land in Ondo state, Nigeria indicated that *Termitomyces mammiformis* was a very good source of crude protein (Adejumo and Awosanya, 2005). They also observed that lipids, sodium and phosphorus contents of the four species were generally very low. Ita *et al.*, (2006) analyzed fruiting bodies of ten mushroom species (edible & non-edible) for their heavy metals contents. Results indicated that the concentration of heavy metals accumulated by the mushrooms were species-dependent. Grillo *et al.*, (2009) evaluated the yield of *Pleurotus pulmonarius* on different mixtures of cotton waste and cassava peel. They found out that cotton waste was a better substrate for cultivation of *P. pulmonarius* than cassava peel. In turkey, Ayhan (2001)

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analyzed six different species of wild mushrooms growing in the east Black Sea region spectrophotometrically for their trace element (Pb, Cd, Hg, Cu, Mn, and Zn) levels. The metal bioaccumulation levels of six mushrooms were high.

Apart from serving as a protein food, mushrooms are also medicinal and have some biotechnology–based functions (Taniguchi, 2000). Mushrooms have been useful in preventing diseases such as hypertension, hypercholesterolemia, cancer and also having antibacterial and antiviral properties. These functional characteristics are mainly due to their chemical composition (Cochran, 1978; Chovot et al., 1997; Gunde-Cinerman, 1999; Manzi *et al.*, 2001). One of the values of commercial cultivation of mushrooms, especially in developing countries like Nigeria, is the availability of large quantities of several agro–industrial wastes which can serve as substrates for the cultivation of mushrooms (Banjo *et al.,* 2004).

It has been reported that mushrooms can grow on cocoa pods, cotton waste, dried chopped maize straw, oil palm wastes, tobacco straw, used tea leaves, rice straw, sugarcane bagasse, newsprint, old rags and sawdust (Banjo *et al.,* 2004). However, very little information is available on mushroom cultivated on waste cardboard which are normally managed in Nigeria through open burning without regards to associated environmental and health risks. Therefore, the focus of this study is to explore alternative management technique for waste cardboard in a sound and environmentally friendly manner, besides beneficial uses for mushrooms grown on its compost.

MATERIALS AND METHODS

Materials used in this study included waste cardboard paper, Cow Intestinal Waste (CIW) and Abattoir Waste Water (AWW). Waste cardboard was sourced from wastes generated at Alesinloye market in Ibadan, Nigeria and both CIW and AWW were collected from the market abattoir. Other materials included simple composting tools like garden fork, spade and weighing scale. Compost thermometer (ts 005110510) and a digital pH meter (rapidest made by Luser Leaf Products Inc., Chaina) were used to monitored the progress of composting. Composting recipe was made by mixing the cardboard and CIW in the ratio 2:1 and periodic wetting was done with AWW. Mature mushroom was harvested at the end of 28 days. Physical observation like odour, colour, type of stem and cap, flesh and spore colour. The results obtained were then compared to those available on internet (Roger, 2012) for proper identification of the mushroom. The mushroom was then uprooted randomly from the compost, air-dried, milled, stored in a labeled black polythene bags and then sent for chemical analysis.

The fruiting bodies of the mushrooms were analyzed for their total organic-carbon (C), total nitrogen (N), C:N ratio, total phosphorus (P), magnesium (Mg), sodium (Na), potassium (K), according to the American Public Health Association (APHA, 2005). The values of iron (Fe), lead (Pb), chromium (Cr), nickel (Ni), copper (Cu), manganese (Mn), zinc (Zn), and cadmium (Cd) contents were read on Atomic Absorption Spectrophotometer after standardizing with respective elements. Windrow composting procedures, laboratory chemical analysis for macro nutrients and selected heavy metal were explained in details in previous study (Hammed *et al.,* 2011). Apart from heavy metal and macronutrient contents that were determined in the samples, proximate analysis was also carried out to assess its suitability for human consumption. Two mushroom species were analyzed for food composition in duplicates, according to the Association of Official Analytical Chemists (AOAC, 1998). The parameters determined were: crude protein, crude fat, moisture content, dry matter, ash, and crude fiber. All analyses were carried out in a soil and fertilizer laboratory, Institute of Agricultural Research and Training (I. A. R. & T.), Moor Plantation, Ibadan.

RESULTS

Physical observation and Taxonomic Characterization of the Mushrooms

Table 1 shows results obtained from the physical observation of the mushrooms. From all observations, the two mushroom species grown naturally from waste cardboard compost were *Inocybe margaritispora and Coprinus narcoticus.* These species are shown in Plates 1-3. In order to ascertain their edibility the two species were not consumed by local hens when thrown to them. Synonymously, the mushrooms are also named as *Sternsporiger Risspilz and Mist-Tintling* respectively. It was not determined whether season had impact on the blossoming of the mushroom. However, from observation, they can be grown throughout the year once necessary conditions are met.

Chemical Characterization of Mushroom for Macro Minerals

Tables 2 and 3 show the results obtained from the chemical characterization of the two types of the mushroom species grown on the paper substrate for their macro minerals. In the two species, organic carbon was found to be 35.58% and 33.28%, nitrogen: 5.58% and 4.90% and phosphorus: 0.33% and 0.24% respectively. In comparing the two types of mushrooms, all the chemical parameters analyzed, with the exception of potassium were found higher in the ***Inocybe margaritispora*** as shown in Figure 1.

Heavy Metal Composition of Mushroom

In order to assess the quality of the mushroom in terms of their toxic potential, levels of some heavy metal were determined in the laboratory. Tables 4 and 5 show the results obtained from this analysis. The levels of these metals varied in two species and with exception to lead and chromium, values of other metals were found higher in *Inocybe margaritispora.* The highly toxic metals with tested adverse health effects like lead and cadmium were found to be 18.35mg/kg and 31.30mg/kg: and, 1.88mg/kg

Table 1 Physical Observation and Characterization of mushrooms

S/N	Indicator	*Inocybe margaritispora*	*Coprinus narcoticus*
1	Fungus colour	White to cream, and Brownish when dried.	White stem and black cap
2	Normal size	Less than 5cm	Less than 5cm
3	Stem type	Simple round stem	Swollen stem
4	Flesh	Very soft which became tough when dried	Granular and brittle when fresh and shrink when dried
5	Spore colour	Light to dark brown	Purplish to black
6	Gills	Whitish at first then clay-brown	White rapidly becoming black,
7	Cap type	Distinctly scaly, especially when dried.	Conical or nearly so. Mealy covering of cap formed of thin-walled cells ornamented with granular warts. The cap later separated into tentacles and became amorphous at maturity which stained object that came near it.
8	Odour	Mushroom has distinct or odd smell (non mushroomy)	Smell strongly of tar-gas

Plate 1 The Two Mushrooms (*Inocybe margaritispora and Coprinus narcoticus*) Growing on Cardboard Compost

Plate 2 *Inocybe margaritispora*

and 1.37mg/kg in *Inocybe margaritispora* and *Coprinus narcoticus* respectively. In addition, some metals that are required by body for proper functioning like iron and zinc were higher in *Inocybe margaritispora* (Figure 2). Results

Plate 3 *Coprinus narcoticus*

Table 2 Chemical Characterization of *Inocybe margaritispora*

Element (%)	*Sample Description*		
	Sample I	Sample II	Mean ± S.D (n=2)
Nitrogen	5.56	5.59	5.58 ± 0.02
Organic carbon	35.31	35.38	35.35 ± 0.04
C: N	5.35	6.33	5.84 ± 0.49
Sodium	0.26	0.28	0.27 ± 0.01
Magnesium	0.56	0.51	0.54 ± 0.03
Calcium	1.89	1.84	1.87 ± 0.03
Phosphorus	0.34	0.31	0.33 ± 0.02
Potassium	3.29	3.26	3.28 ± 0.02

Table 3 Chemical Characterization of *Coprinus narcoticus*

Element (%)	***Sample Description***		
	Sample I	**Sample II**	**Mean ±S.D (n=2)**
Nitrogen	4.89	4.91	4.90 ± 0.01
Organic carbon	33.28	33.28	33.28 ± 0.00
C: N	6.81	6.78	6.80 ± 0.02
Sodium	0.15	0.17	0.16 ± 0.01
Magnesium	0.44	0.48	0.46 ± 0.02
Calcium	1.46	1.50	1.48 ± 0.02
Phosphorus	0.25	0.23	0.24 ± 0.01
Potassium	3.16	3.15	3.16 ± 0.01

of Proximate Analysis of the *Inocybe margaritispora* and *Coprinus narcoticus*

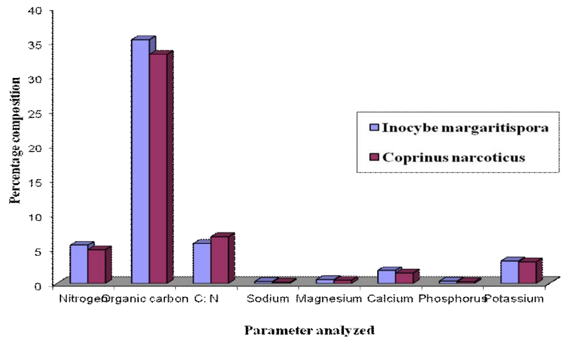

Fig.1 Comparison of Macro Element in the two mushroom species

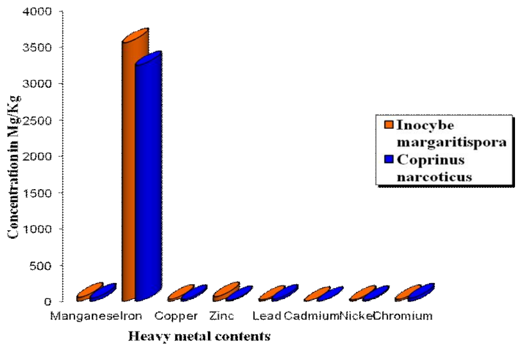

Fig.2 Comparison of Heavy Metals in the Two Samples of Mushroom

Table 4 Heavy Metals Composition of *Inocybe margaritispora*

Parameter (mg/kg)	*Sample Description*		
	Sample I	**Sample II**	**Mean ± S.D (n=2)**
Manganese	58.70	58.30	58.50 ± 0.20
Iron	3562.00	3560.00	3561.00 ± 1.00
Copper	31.00	30.50	30.75 ± 0.25
Zinc	67.00	66.50	66.75 ±0.25
Lead	18.30	18.40	18.35 ± 0.05
Cadmium	1.90	1.85	1.88 ± 0.03
Nickel	17.30	17.40	17.35 ± 0.05
Chromium	26.70	26.50	26.6 ± 0.10

Table 5 Heavy Metals Composition of Coprinus narcoticus

Parameter (mg/kg)	Sample Description		
	Sample I	Sample II	Mean ± S.D (n=2)
Manganese	46.30	46.50	46.40 ± 0.01
Iron	3249.00	3249.00	3249.00 ± 0.00
Copper	26.00	25.50	25.75 ± 0.25
Zinc	58.00	59.00	11.70 ± 0.50
Lead	15.50	15.80	31.30 ± 0.15
Cadmium	1.30	1.23	1.37 ± 0.04
Nickel	14.20	14.60	14.40 ± 0.20
Chromium	23.40	23.50	46.90 ± 0.05

Table 6 Proximate Analysis of *Inocybe margaritispora* and *Coprinus narcoticus*

Parameter Determined (%)	*Coprinusnarcoticus*			*Inocybemargaritispora*		
	Sample I	**Sample II**	**Mean ± S.D (n=2)**	**Sample I**	**Sample II**	**Mean ± S.D (n=2)**
Crude protein	26.38	26.32	26.35 ± 0.03	29.52	29.57	29.55 ± 0.03
Crude fat	13.41	13.37	13.39 ± 0.02	15.38	15.41	15.40 ± 0.03
Crude fibre	11.75	11.79	11.77 ± 0.02	13.47	13.44	13.46 ± 0.02
Dry matter	88.12	88.14	88.13 ± 0.01	88.56	88.51	88.54 ± 0.03
Ash	12.36	12.34	12.35 ± 0.01	14.28	14.30	14.29 ± 0.01

The parameters tested for nutritional values of the mushrooms were: *percentage crude fibre, crude fat, protein, dry matter and ash.* In all parameters, percentage dry matter was the highest in the two samples. This is followed by the percentage crude protein and percentage crude fat. It was observed that all the parameters were higher in *Inocybe margaritispora.*

DISCUSSIONS

Chemical Characterization of Mushroom for Macro Minerals

Various studies have shown that mushrooms can grow in a highly cellulose organic wastes (Banjo et al., 2004). In the study, mushroom was cultivated on waste cardboard paper. The natural phenomenon that surrounds the growth of mushroom on these wastes could be traced to their capability to degrade lignin. At the extinction of the toughest composting bacteria during the aerobic decomposition of high cellulose organic wastes, such wastes are quickly colonized by fungi species which feed on the dead bacterial and break down the leftover waste for complete stabilization. The results of mineral values of the two species of mushrooms clearly indicated their potentials as good sources for nitrogen and potassium. The vast nitrogen content could have resulted into the high levels of percentage crude protein. The results of valuable minerals like calcium, sodium and phosphorus are comparatively lower and not in agreement with the report of analysis of some cultivated mushrooms like *Agaricus bisporus*, *Lentinus edodes*, and *Pleurotus ostreatus* (Mattila et al., 2001). They are also lower than those reported for several cowpea varieties (Aletor and Aladetimi, 1989) and those reported for fish, snails and broiler meat (Imevbore, 1992). However, lager values were obtained for iron and calcium that are also required for proper functioning of body systems.

Heavy Metal Composition of Mushroom

The higher values of heavy metals in the mushrooms which exceeded the stipulated FAO/WHO (1976) dietary standards could be traced to numerous coating, staple pins, sizing agents, colourants and other printing materials used in cardboard production. The results are not in consonance with the trend: Fe > Zn > Cu > Mn > Pb > Cd observed by Ita *et al* (2006). The values of iron obtained in this study were far higher than what they obtained for *Polyporus frondosis* with a maximum value of 731.6 ± 13.2 mg kg-1. Kuusi *et al* (1981) and Kalac and Svoboda (2000) showed that mushrooms could build up larger concentrations of some heavy metals particularly cadmium (Cd^{2+}), mercury (Hg^{2+}) and lead (Pb^{2+}) when compared to green plants. This suggests that mushrooms possess a very effective mechanism that enables them readily to take up heavy metals from the substrates (Turkekul et al., 2004). Kalac and Svoboda (2000) reported that the age of the fungal fruiting body or its size is of less importance in the accumulation of heavy metals by mushrooms.

Zinc is widespread in living organisms due to its biological significance. The level reported here for the *Inocybe margaritispora* (66.75 ±0.25) falls within the ranges values reported by Turkekul *et al.,* 2004; Tuzen et *al.,* 1998 and Kalac and Svoboda, 2000. Isildak *et al.,* (2004) reported a Cu concentration of 107 ±8.5 µg g^{-1} in wild growing *Agaricus biosporus* from the middle black sea region of Turkey which was far higher that the values obtained for the two mushrooms analyzed in this study. However, the Cu range obtained in this study is in agreement with reported range of 10 – 70 µg g^{-1} (Anderson *et al.,* 1982; Vetter, 1994; Agrahar-Murugkar & Subbulakshmi, 2005). The levels of Pb, Cd, and Mn were found to be higher than the values obtained in the previous studies (Falandysz and Bona, 1992; Vetter 1994; Ayhan 2001; Ita *et al.,* 2006) and international standard (Food Sanitation Standards, 2007). The Fe values obtained are similar to those values obtained in other studies: Latiff *et al.,* (1996) reported Fe concentration range of 100 – 1216 µg g^{-1} in mushroom and Turkekul *et al.,* (2004) reported Fe content of 568 – 3562 mg kg^{-1} DM in mushroom samples from Tokat, Turkey. Variations in Fe content may be attributed to the different mushroom species, uptake levels and the levels accumulated by the substrate from, which the mushrooms were harvested (Ita *et al.,* 2006).

Proximate Analysis of the *Inocybemargaritispora* and *Coprinusnarcoticus*

Results of proximate analysis indicated that *Inocybe margaritispora* was a very good source of crude protein (59.05%). This is higher than what was obtained in a previous study conducted by Adejumo and Awosanya (2005) where 37% of crude protein was found. The levels of crude fibre ash are also higher than what was obtained by Adejumo and Awosanya (2005). It is, however, similar to the finding of Miles and Chang (1986 and 1997). Ogundana and Fagade (1981) indicated that mushroom was about 16.5% dry matter out of which 7.4% was crude fiber, 14.6% was crude protein and 4.48% was fat and oil. The values obtained for the respective parameters were higher in this study. According to Jiskani (2001), the protein value of mushrooms is twice as that of asparagus and potatoes, four times as that of tomatoes and carrots, and six times as that of oranges. The ash and fiber contents were higher than the ones in previous studies (Aletor and Aladetimi, 1995). The author reported 3.7% fiber and 13.9% ash contents for *T. robustus.* The crude protein, ash and crude fiber values of most mushrooms compared favourably with and in some instances surpassed those reported for most legumes except groundnut and soybeans grown in West Africa (FAO, 1970; Aletor and Aladetimi, 1989). In addition, apart from serving as a protein food, for which they are most popular in Nigeria, mushrooms are also medicinal and have some biotechnology–based functions (Taniguchi, 2000).

CONCLUSIONS

It was evident in this study that waste cardboard paper could be used to cultivate mushroom. The species that colonized composted waste cardboard at maturity were *Inocybe margaritispora* and *Coprinus narcoticus*. Although the two species were found to contain high levels of nutritional parameters like crude protein, crude fibre and crude fat- which were more in *Inocybe margaritispora*, that could have qualify them as edible mushroom species, due to the high levels of heavy metals and lower macro nutrients like sodium and magnesium, it is not advisable to utilize them for human consumption. However, they can find use in other applications in medicine and some other biotechnology–based functions. Other studies are required to harvest the spores of these mushroom species, especially that of *Inocybe margaritispora* with higher potential for heavy metal accumulation. The spore can be cultivated on land polluted with heavy metal for the purpose of bio-remediation and soil reclamation. It is therefore, recommended that mushroom should be analyzed for heavy metal contents before processing for human consumption because of their high level of heavy metal accumulation.

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