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Antifungal effect of *Coriolopsis polyzona* (Pers) on fungi isolated from remnant foods and wastewater from restaurants in Akure metropolis, Nigeria

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The present study assessed the antifungal properties of extracts of *Coriolopsis polyzona* against fungi isolated from remnant foods and wastewater that are discharged into the environment without treatment. Species of fungi isolated from remnant foods and wastewater in Akure metropolis are *Aspergillus fumigatus*, *Aspergillus niger*, *Mucor mucedo*, *Penicillium chrysogenum*, *Apsergillus flavus*, *Fusarium oxysporum*, *Penicillium italicium*, *Triscelophorus monosporus* and *Rhizopous stolonifer*. Phytochemical constituents in the extracts of *C. polyzona* ranged from 0.75 to 14.20 mg/g for saponins, flavonoids, alkaloids, tannins and steroids. The percentage inhibition of fungal mycelia by *C. polyzona* extracts against isolated fungi at concentration of 50 to150 mg/ml ranged from 2.7 to 37.7%. Purified extracts exhibited better mycelia inhibition of 42.2% on *Penicillium chrysogenum* and 34.1% on *Aspergillus fumigatus* that were resistance to fluconazole and ketoconazole respectively. The presence of phytochemicals and functional groups such as hydroxyl (OH), alkyl (C-H) carbonyl (C=O) and aromatic ring (C=C) in the extracts of *C. polyzona* may be responsible for the antifungal effect of the extracts. Compounds associated with these functional groups if isolated and purified may help in combating the present phenomenon of resistance of fungi to commercial antifungal agents.

Key words: Restaurant wastes, *Coriolopsis polyzona*, mycelia inhibition, functional groups, environment.

INTRODUCTION

Mushrooms are medicinal foods used extensively in traditional medicine for combating diet-related diseases, treating chronic diseases, delay ageing and increase life expectancy (Khatun et al., 2012). Mushrooms' medicinal properties are owing to the possession of wide range of secondary metabolites that are of high therapeutic values. Some of these medicinal properties are antibacterial, antifungal, antiviral, anticancer, anti-inflammatory and antioxidants (Wasser, 2002; Ameri et al., 2011; Keles et al., 2011). Moreover, the combination of vitamins, essential mineral salts, protein, polysaccharides, fibres and low lipid content in mushrooms are dietary supplements to strengthen the immune system (Wani et al., 2010). Species of *Termitomyces*, *Ganoderma, Pleurotus, Cordyceps sinensis, Inonotus obliquus, Polyporus umbellatus, Shizophyllum commune* and others

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are medicinal macrofungi with nutraceutical properties (Barros et al., 2008; Jiang and Sliva, 2010; and Hesham et al., 2013).

The nutraceutical activities of medicinal macrofungi had been traced to the presence of phytochemicals. The biological and metabolic diversity of mushrooms are due to their ability to utilize several substrates; decomposing dead organic matters and thus explore different habitat for colonization (Thatoi and Singdevsachan, 2014). This bioconversion of organic waste materials has not only brought about biodegradation but plays a major role in nutrient cycles that are of great benefits to man and nature. *Coriolopsis polyzona* is a white rot fungus commonly found on decaying woods and known to mineralize lignin and phenolic compounds by their multienzyme systems (Alaoui et al., 2008). The bio-assimilation of higher molecular compounds by macrofungi has led to the secretion of biological active compounds that are of great medicinal importance. However, the continuous searching for novel bioactive compounds in medicinal mushrooms will yield greater success in pharmaceuticals and food science. This will definitely provide perpetual solution to the significant increase in antibiotics resistance by pathogenic organisms and continuous failure witnessed in some chemotherapy agents.

Restaurants in Akure metropolis dispose their wastewater and remnant foods to the environment without treatment. These wastes are laden with pathogenic organisms that are resistant to antibiotics (Ogidi and Oyetayo, 2013). The indiscriminate dumping of refuse containing pathogenic organisms into the environment had contributed to the transfer of resistance genes in the ecosystem, contamination of watercourses and causes of eutrophication (Akpor and Muchie, 2011).

Fungi spores are widely dispersed into air and these spores are known to contribute to allergic diseases (Hageskal et al., 2009). Brakhage et al. (2010) reported that fungi produce volatile organic compounds that contribute to ill health. Therefore the emerging and transfer of resistance genes by pathogenic fungi containing spores in food chain cannot be overemphasized. This situation has contributed to the ineffectiveness of antibiotics. The search for effective antifungal agents had increased recently. Attention had been turned to mushrooms because they are known to contain bioactive compounds that are effective against microbial entities (Lindequist et al., 2005). The present study is therefore aimed at assessing the antifungal property of extracts of *Coriolopsis polyzona*, a wild macrofungus, against fungi isolated from wastewater and remnant foods.

MATERIALS AND METHODS

Collection of macrofungus

Samples of macrofungus were collected from farmland and nearby forest at the Federal University of Technology Akure, Nigeria (Lat 07° 14^{l}N Long 05° 11^{l}E). Sample of fruiting body was morphologically identified and confirmed by molecular methods.

Preparation of extracts from *C. polyzona*

Collected samples of *C. polyzona* were dried and ground to powder using mill machine (Retsch GmbH 5657 HAAN). The fine powder of *C. polyzona,* 100 g each, was soaked in acetone and methanol for 48 h. The filtrates obtained were dried using rotary evaporator (RE-52A, UNION Laboratories, England) and labelled as ACE and MCE for acetone extract and methanol extract respectively.

Isolation and Identification of fungi

Samples of wastewater and remnant foods were examined microbiologically using standard methods. The microscopic identification was done according to Chander (2002) and Barnett et al. (1983). The identified fungi were maintained on Potato dextrose agar (PDA Lab M) slants at 4°Cin refrigerator for subsequent used.

Quantitative determination of phytochemical contents in extracts of *C. polyzona*

The flavonoid content was determined by aluminum chloride colorimetric method described by Singleton et al. (1999). The methods described by Harborne (1998) were adopted to quantify alkaloid content of the extracts while tannins, saponins, tepernoids and steroid amount was quantified using the method described by AOAC (2003).

Antifungal Activities of the crude extracts and purified fractions

The antifungal activities of extracts were determined by Poison food technique (Parajuli et al., 2005). Volume of 1.0 ml each concentration (50, 100 150 mg/ml) of acetone extract (ACE) and methanol extract (MCE) were aseptically poured into Petri dish followed by the addition of equal amount of PDA. The Petri dish was agitated while adding PDA so as to get even mixture of the contents. PDA plates with dimethyl sulfoxide (DMSO) serves as control. Seven days old culture of the test fungi were used to prepare inoculums discs (6mm diameter) using cork borer. Fungal inoculum was aseptically placed upside down in the centre of each plate containing MCE and ACE. The experiment was performed in three replicates. The average diameter of fungal colonies was measured on the 7th day after inoculation and percentage of mycelia growth inhibition was calculated. The Percentage growth inhibitions in different concentrations were calculated as:

$$\text{Percentage growth inhibition} = \frac{g_c - g_t}{g_c} \; x \; 100$$

Where, g_c = Growth of mycelia colony after incubation period in control set subtracting the diameter of inoculums disc; g_t = Growth of mycelia colony after incubation period in treatment set subtracting the diameter of inoculums disc.

The antifungal effect of partially purified fractions was also carried out according to the method above.

Partial purification of extracts

The separation of extracts was done according to the method of Owolabi and Olarinoye (2008). The column chromatography was

Table 1 .Fungal isolates from sampled remnant foods and wastewater from restaurants in Akure metropolis.

Fungi Isolates	RMF	WWS
Aspergillus fumigatus	+	+
Aspergillus niger	+	+
Mucor mucedo	+	–
Penicillium chrysogenum	+	+
Apsergillus flavus	+	–
Fusarium oxysporum	+	+
Penicillium italicium	+	+
Triscelophorus monosporus	–	+
Rhizopous stolonifer	+	–

RMF= Remnant foods; WWS= wastewater; + = present; – = absent.

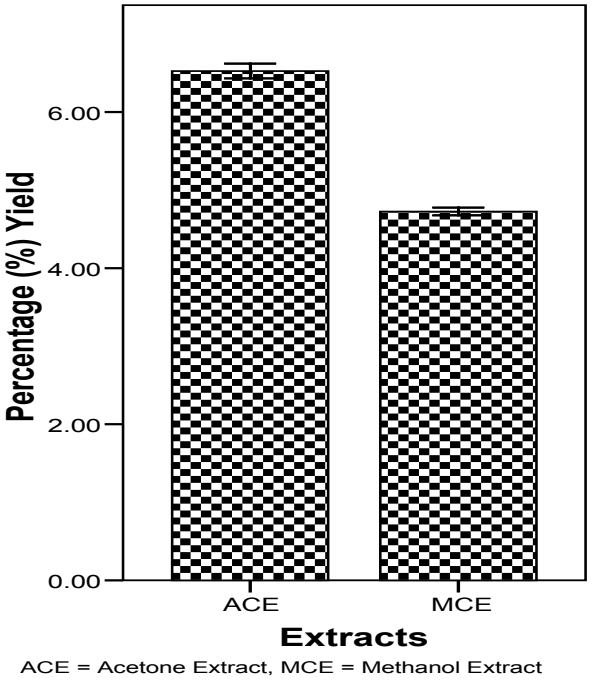

Figure 1. Percentage yield of acetone and methanol extracts of *Coriolopsis polyzona.*

Table 2. Quantitative constituents of phytochemicals in extracts (mg/g) of *Coriolopsis polyzona.*

Phytochemicals	ACE	MCE
Saponins	2.10±0.00	1.50±0.11
Flavonoids	6.80±0.04	4.55±0.02
Alkaloids	14.20±0.10	11.60±0.04
Tannins	5.20±0.04	7.70±0.20
Steroid	0.75±0.01	–

Values are mean±SD of replicates. ACE= acetone extract; MCE= methanol extract.

filled with petroleum ether (500ml), 60 grams of silica gel (60-120mesh) was tamped into column chromatography. The sample of extracts was mixed with silica gel and ground together to homogenize. This was transferred with spatula into the packed column chromatography.The column was opened and eluants were collected in 100ml aliquot portion. The polarity of mobile phase was varied by introducing chloroform and methanol. Thin layer chromatography (TLC) plate was saturated in chromatographic tank containing chloroform: methanol (10: 5) solvent systems.

Infra-red spectroscopic analysis

The presence of functional groups in *Coriolopsis polyzona* extracts was assessed using FT Infra-Red spectrophotometer (Spectrum BX).The FT IR wave number (cm^{-1}) obtained was interpreted according to Willams (1982).

Statistical analysis

The experiment was carried out in triplicates. Data obtained were analyzed using one way analysis of variance and means were compared by Duncan's Multiple Range Test using (SPSS 17.0 version).

RESULTS

Species of fungi isolated from remnant foods and wastewater collected from restaurants are shown in Table 1. The fungi isolated are *Aspergillus fumigatus*, *Aspergillus niger*, *Mucor mucedo*, *Penicillium chrysogenum*, *Apsergillus flavus*, *Fusarium oxysporum*, and *Penicillium italicium.* Figure 1 shows the percentage yield of methanol and acetone extracts. Acetone has the highest percentage yield (6.5%) than methanol (4.7%). Table 2 shows the phytochemical contents of *C. polyzona* extracts. Acetone extract (ACE) had alkaloid content of 14.20 mg/g while methanol extract (MCE) had 11.6 mg/g. Flavonoids content of the extracts ranged from 4.55 to 6.80 mg/g and tannins is 5.2 and 7.7mg/g for ACE and MCE respectively.

The mycelia inhibition of *C. polyzona* extracts ranged from 4.7 to 37.7% against isolated fungi from remnant foods and 2.7 to 37.3% against isolated fungi from wastewater samples at different concentration of 50, 100 and 150 mg/ml (Tables 3 and 4). Acetone and methanol extracts of *C. polyzona* exhibited highest mycelia inhibition against *Penicillium chrysogenum* and *A. niger* than mycelia inhibition caused by fluconazole (Table 3). Acetone extract shows mycelia inhibition of 37.7% against *P. chrysogenum* that was found to show resistance against Ketoconazole (Table 4). Plate 1a shows the mycelia inhibition of *C. polyzona* against isolated fungi while Plate 1b containing DMSO (control) shows no mycelia inhibition against tested fungi. The percentage of mycelia inhibition observed in purified extracts is closely related to the commercial antibiotics used as positive control (Tables 3, 4 and 5). The purified fractions of *C. polyzona* also exhibited more potent mycelia inhibition against *P. chrysogenum*, *A. fumigatus*,

Table 3. Percentage of mycelia inhibition of fungi isolates from remnant foodsby*Coriolopsis polyzona* extractsat different concentrations (mg/ml).

Tested isolates	MCE 150	ACE 150	MCE 100	ACE 100	MCE 50	ACE 50	KET 20	FLU 20
Penicillium italicium	17.3±0.5	21.6±0.4	14.8±0.1	10.3±0.1	5.9±0.0	6.4±0.1	56.2±0.1	0.0±0.0
A. niger	27.9±0.9	24.8±0.2	20.8±0.0	17.0±0.0	0.0±0.0	11.8±0.0	41.0±0.0	29.0±0.0
Fusarium. oxysporum	12.4±0.5	26.4±0.4	10.6±0.5	12.3±0.0	9.1±0.0	8.4±0.0	61.6±0.0	75.6±0.0
Mucor mucedo	26.6±0.4	22.4±0.7	13.6±1.1	20.9±0.0	9.0±0.0	8.8±0.0	42.8±0.2	62.4±0.1
Rhizopous stolonifer	0.0±0.0	8.9±0.0	0.0±0.0	0.0±0.0	0.0±0.0	0.0±0.0	68.2±0.1	38.0±1.0
Penicilliumchrysogenum	28.5±0.9	37.7±0.1	15.8±0.1	24.5±0.0	10.2±0.0	12.4±0.1	50.5±0.0	50.6±0.5
A. fumigatus	20.3±0.3	26.6±0.5	10.3±0.6	21.2±0.0	0.0±0.0	10.0±0.0	59.7±0.0	0.0±0.0
A. flavus	0.0±0.0	10.7±0.2	0.0±0.0	7.6±0.0	0.0±0.0	4.7±0.1	68.0±0.0	53.7±0.3

Values are mean of replicates (n = 3). 0.0 = no mycelia inhibition; ACE= acetone extract; MCE= methanol extract. FLU= fluconazole; KET=ketoconazole.

Table 4. Percentage of mycelia inhibition of fungi isolates from wastewater by *Coriolopsis polyzona* extractsat different concentrations (mg/ml).

Tested isolates	MCE 150	ACE 150	MCE 100	ACE 100	MCE 50	ACE 50	FLU 20	KET 20
Fusarium oxysporum	5.6±0.0	8.0±0.0	0.0±0.0	4.0±0.0	0.0±0.0	4.0±0.3	56.6±0.5	68.0±0.0
Penicullium italicium	2.6±0.0	10.7±0.1	0.0±0.0	0.0±0.0	0.0±0.0	0.0±0.0	64.0±0.0	63.6±0.1
Asperillus fumigatus	10.3±0.2	24.2±0.0	5.0±0.0	8.3±0.0	2.7±0.0	2.7±0.0	0.0±0.0	40.8±0.1
Triscelophorus monosporus	10.5±0.1	21.2±0.3	5.9±0.1	10.4±0.0	4.7±0.0	4.0±0.0	61.5±0.0	59.4±0.1
Penicillium chyrsogenum	12.6±0.1	37.3±0.2	5.0±0.0	20.1±0.0	5.4±0.0	8.0±0.0	32.5±0.0	0.0±0.0
Aspergillus niger	10.3±0.0	7.9±0.0	.0±0.0	0.0±0.0	0.0±0.0	0.0±0.0	21.7±0.1	41.7±0.1

Values are mean of replicates (n = 3). 0.0 = no mycelia inhibition; ACE= acetone extract; MCE= methanol extract. FLU= fluconazole; KET=ketoconazole.

Plate 1a. Mycelia inhibitionof *Aspergillus niger* by*Coriolopsis polyzona* extract.

Plate 1b. Negative control.

F. oxysporum and *A. niger* as shown in Table 5. Table 6 shows the functional groups of bioactive compounds in *C. polyzona* extracts. The results of FTIR analysis revealed the presence of the following functional groups; hydroxyl (OH), alkyl (C-H), carbonyl (C=O) and aromatic ring (C=C).

Table 5. Mycelia Inhibition (%) of purified fractions of *Coriolopsis polyzona* against isolated fungi at 5.0 mg/ml.

Parameter	AC	AP	MC
Isolated fungi from remnant foods			
Penicillium italicium	25.0	22.4	ND
A. niger	30.3	0.0	ND
Fusarium. oxysporum	32.2	31.2	ND
Mucor mucedo	27.8	23.5	ND
Rhizopous stolonifer	17.5	20.5	0.0
Penicilliumchrysogenum	40.6	42.2	38.3
A. fumigatus	34.1	0.0	23.8
A. flavus	15.6	0.0	0.0
Isolated fungi from wastewater			
Fusarium oxysporum	13.3	15.3	ND
Penicullium italicium	8.8	11.2	ND
Asperillus fumigatus	19.4	20.7	ND
Triscelophorus monosporus	22.5	ND	ND
Penicillium chyrsogenum	15.2	ND	ND
Aspergillus niger	14.3	14.1	ND

Values are mean of replicates (n = 3). AC= purified fraction from acetone extract; AP= purified fraction from acetone extract; MC= purified fraction from methanol extract; ND= Not detected.

Table 6. Functional groups obtained from purified fractions of *Coriolopsis polyzona* at different wave number (cm^{-1}).

IRWave Number (cm^{-1})	**Functional groups**
AC	
3377.3	OH stretch
2945.15	CH_3/CH stretch
2834.78	CH_3/CH stretch
1652.27	C = C stretch
1410.30	C=O stretch
1026.12	C – O stretch
AP	
3428.33	OH stretch
2932.99	CH stretch
1637.26	C=C stretch
1405.11	C=O stretch
1030.85	C – O stretch
MC	
3426.00	OH stretch
1642.12	C=C stretch
1399.65	CH stretch
1021.65	C –O stretch

AC= purified fraction from acetone extract;AP= purified fraction from acetone extract;MC= purified fraction from methanol extract.

DISCUSSION

The practice of disposing wastes from restaurants without treatment had been in existence without check in most communities in Nigeria. This has led to build up of chemical and biological pollutants in the environment. The accumulation of biological pollutants such as microorganisms in the environment beyond self-purification will be a threat to public health (Akpor and Muchie, 2011). Build-up of microorganisms in the environment has also contributed to the problem of resistance of microorganisms to commercial antibiotics as a result of exchange of resistant genes (Frost et al., 2005). This research work therefore assessed the antifungal effect of extracts obtained from a wild macrofungus, *C. polyzona,* against fungi isolated from restaurant wastes.

The genera of fungi isolated from remnant foods and wastewater from restaurants in Akure metropolis includes *Aspergillus*, *Penicullium*and *Fusarium.* The occurrence of different species of fungicould be due to the exposure time of food during processing and after processing to airborne fungi, the microbiological quality of water used for food preparation, sanitary quality of the equipment and health status of employees. The importance of these fungi to endanger human health had been highlighted (Warris et al., 2001). Hedayati et al. (2011) had earlier isolated these groups of fungi from hospital drinking water in Iran. The findings of Oranusi et al. (2011) revealed that some of these fungi are of public health importance. Makun et al. (2009) also reported that many of these fungi species produce toxic metabolites in Nigerian staples and ascribed the mycotoxins to species of *Aspergillus*, *Penicillium*, *Fusarium* and *Mucor*.

The phytochemical contents in extracts of *C. polyzona* are flavonoids, alkaloids, tannins, saponins and steroids.

These phytochemicals had been reported to possess antifungal effects (De Silva et al., 2013). Therefore, the inhibitory potentials of extracts obtained from *C. polyzona* were well pronounced against isolated fungi from waste-water and remnant foods (Tables 2, 3 and 4). *Coriolopsis* species had earlier been reported to possess antimicrobial properties (Oyetayo et al., 2010). Gbolagade et al. (2007) had also earlier reported the antifungal properties of some Nigerian higher fungi on species of pathogenic fungi.

The percentage inhibition of mycelial growth by extracts of *C. polyzona* was similar to the antifungal activities of some wild macrofungi reported by Imtiaj and Lee (2007).The mycelia inhibition (2.7 to 37.3%) caused by extracts of *C. polyzona* on the isolated fungi conformed to results obtained by Ayodele and Idoko (2011) who reported mycelia inhibition of 13.82 to 32.10% against *A. niger* and *Penicillium notatum* by culture filtrate of *Lentinus squarrosulus, Psathyrella atroumbonata, Volvorella volvacea* and *Coprinellus micaceus*.

The mycelia inhibition of these isolated fungi by *C. polyzona* extracts could be attributed to the presence of secondary metabolites, which comprises of different functional groups such as ketones, hydroxyl, methyl, carboxylic acid and aromatic ring. Compounds possessing these functional groups are known to be potentially useful phytochemicals that can exhibit antimicrobial activities (Cowan, 1999). Barros et al. (2008) and Rai et al. (2005) had associated the medicinal values of macrofungi to diversities of secondary metabolites that contained different functional groups of biologically active compounds.

The possession of antifungal properties by extracts of *C. polyzona* show that it contains bioactive compounds that could be extracted and explored as new drugs against the continuous failure of chemotherapies and prevalent resistance of microorganisms to commonly used antibiotics.

Conflict of interests

The authors did not declare any conflict of interest.

REFERENCES

Akpor OB,Muchie M (2011). Environmental and public health implications of wastewater quality. Afr. J. Biotechnol. 10(13): 2379-2387.

Alaoui SM., Merzouki M, Penninckx MJ, Benlemlih M (2008). Relationship between cultivation mode of white rot fungi and their efficiency for olive oil mill wastewaters treatment. Electronic J. Biotechnol. 11(4):1-8.

Ameri A, Ghadge C, Vaidya JG, Deokule SS (2011). Anti-Staphylococcus aureus activity of *Pisolithus albus* from Pune, India. J. Med. Plants Res. 5(4): 527-532.

Association of Official Analytical Chemists (AOAC) (2003). Official Methods of Analysis of the Association of Official Analytical Chemists 17th ed. AOAC International: Gaithersburg,USA.

Ayodele SM, Idoko ME (2011). Antimicrobial activities of four wild edible mushrooms in Nigeria. Int. J. Sci. Nat. 2(1): 55-58

Barnett JA, Payme RW, YawoD (1983). Yeast characteristics and identification, 2nd edition Cambridge University Press, New York.

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

Brakhage AA, Bruns S, Thywissen A, ZipfelPF, Behnsen J (2010). Interaction of phagocytes with filamentous fungi. Curr. Opin. Microbiol. 13:409-415

Chander J (2002). Routine mycological techniques. Medical Mycology. 2nd Edition, Mehta Publishers, New Delhi. pp. 286-398.

Cowan MM (1999). Plant products as antimicrobial agent. Clin. Microbiol. Rev. 12(4):564-582.

De Silva DD,Rapior S, Sudarman E, Stadler M,Xu J,Alias SA, Hyde KD (2013). Bioactive metabolites from macrofungi: ethnopharmacology, biological activities and chemistry. Fungal Diver. 62:1-40

Frost LS, Leplae R, Summer AO, Tonssaint A (2005). Mobile genetic elements: The agent of open source evolution. Nat. Rev. Microbiol. 3:722- 732.

Gbolagade J, Kigigha L, Ohimain E (2007). Antagonistic effect of extracts of some Nigerian higher fungi against selected pathogenic microorganisms. Am. Eurasian J. Agric. Environ. Sci. 2 (4): 364-368.

Hageskal G, Lima N, Skaar I (2009). The study of fungi in drinking water. Mycol. Res. 113:165-172.

Harborne JB (1998). Phytochemical methods. 3rd edition., Chapman and Hall Ltd., London, pp. 135- 203.

Hedayati MT, Mayahi S, Movahedi M, Shokohi T (2011). Study on fungal flora of tap water as a potential reservoir of fungi in hospitals in Sari city, Iran. J. Med. Mycol. 21:10-14.

Hesham EE, Elsayed EA, Aziz R, Wadaan MA (2013). Mushrooms and truffles: historical biofactories for complementary medicine in Africa and in the middle east. Evid. Based Complement. Alternat. Med. 2013:620451.

Imtiaj A, Lee TS (2007). Screening of antibacterial and antifungal activities from Korean wild mushroom. World J. Agric. Sci. 3(3):316-321.

Jiang J, Sliva D (2010). Novel medicinal mushroom blend suppresses growth and invasiveness of human breast cancer cells. Int. J. Oncol. 37(6):1529-1536.

Keles A, Koca I, Gencelep H (2011). Antioxidant properties of wild edible mushrooms. J. Food Prot. Technol. 2 (6): 130-135.

Khatun S, Islam A, Cakilcioglu U, Chatterjee NC (2012). Research on mushroom as a potential source of nutraceuticals: A review on Indian perspective. Am. J. Exp. Agric. 2(1):47-73.

Lindequist U, Niedermeyer TNJ, Julich WD (2005). The pharmacological potential of mushrooms. Evid. Based Complement. Alternat. Med. 2(3): 285-299.

Makun HA, Gbodi TA, Akanya OH, Saloko AE, Ogbadu GH (2009). Health implication of toxigenic fungi found in two Nigerian staples: guinea corn and rice. Afr. J. Food Sci. 3(9):250-256.

Ogidi CO,Oyetayo VO (2013). Antibiotic sensitivity pattern of microorganisms isolated from remnant foods and wastewater from restaurants. FUTA J. Res. Sci. 9 (2):209-216.

Oranusi S, Omagbemi F, Eni, AO (2011). Microbiological safety evaluation of snacks sold in fast food shops in Ota, Ogun state, Nigeria. Int. J. Agric. Food Sci. 1(4): 75-79.

Owolabi BJ, Olarinoye NO (2008). Organic chemistry; The fundamentals. Bohan press, Ibadan. pp. 72-76.

Oyetayo VO, Okuo MO, Olatunji EK (2010). Inhibitory potential of ethanolic extract of two wild polyporales macrofungi on aerobic microbial isolates of meat. Nig. J. Microbiol. 24(1):2033-2037.

Parajuli RR, Tiwari RD, Chaudhary RP, Gupta VN (2005). Fungitoxicity of the essential oils and some aromatic plants of manang against *Alternaria brassicicola*. Sci. World 3(3):39-43.

Rai M, Tidke G, Wasser SP (2005). Therapeutic potential of mushrooms. Nat. Prod. Rad. 4(4): 246-257.

Singleton VL, Orthofer R, Lamuela-Raventos RM (1999). Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Cioalteau Reagents. Methods Enzymol. 299: 152-178.

Thatoi HN,Singdevsachan SK (2014). Diversity, nutritional composition and medicinal potential of Indian mushrooms: A review. Afr. J. Biotechnol. 13(4):523-545.

Wani BA, Bodha, RH, Wani AH (2010). Nutritional and medicinal importance of mushrooms. J. Med. Plants Res. 4(24):2598-2604

Warris A, Gaustad P, Meis JF, Voss A, Verweij PE, Brahamsen TG (2001) Recovery of filamentous fungi from water in a paediatric bone marrow transplantation unit. J. Hosp. Infect. 47:143-148.
Wasser SP (2002). Medicinal mushrooms as a source of antitumor and immunodulating polysaccharides. Appl. Microbiol. Biotechnol. 60:258-274.
Willams DHA (1982). Spectroscopic method in organic chemistry,Willey and Son, London.