

Antimicrobial Activities Of Some Selected Edible Mushrooms And Spices Against Clinical Isolates From Federal University Teaching Hospital Abakaliki (FETHA), Ebonyi State, Nigeria.

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Abstract: The rampant multi-drug resistance among human pathogenic microorganisms has necessitated a continuous search for new and potent antimicrobial substances, especially among plants. Also, the importance of herbal plants as sources of alternative medicine is documented worldwide. In this study, antimicrobial activities of extracts of seven edible mushrooms and two spices (ginger and garlic) against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Streptococcus pyogenes*, *Escherichia coli*, and *Candida albicans* from Federal University Teaching Hospital, Abakaliki (FETHA), Ebonyi State, Nigeria were investigated. Antimicrobial components from the mushrooms and spices were extracted with hot water and cold water, ethanol and diethyl ether; the antimicrobial activities were examined by agar well diffusion method. Zones of inhibition were seen mostly in hot water extracts of five mushrooms (*Trichaptum* sp, *Flammulina* sp, *Boletus* sp, *Tricholoma* sp, and *Psalliota campestris*) on culture plates inoculated with *S. aureus*, *P. aeruginosa*, *S. Pyogenes*, *E. coli* and *C. albicans* at 37°C within 24hrs. The cold water extracts of the mushroom, *P. campestris*, gave the highest zone of inhibition of 25.0 mm, followed by 15.0 mm when used against *P. aeruginosa*. While for spices, the cold water extracts yielded the highest zones of inhibition of 25.0 mm followed by 20.0mm as were observed with ginger. The results obtained have shown clearly that the mushrooms (*Trichaptum* sp, *Tricholoma nudum*, *Psalliota campestris*, *Flammulina* sp, *Boletus* sp), garlic (*Allium sativum*) and ginger (*Zingiber officinale*) extracts contain phytochemicals with some antimicrobial activities while *Cortinarius* sp showed no antimicrobial activity. The water extracts of the mushrooms and spices showed broad spectrum antimicrobial activity much more than ethanol and diethyl ether extracts. Generally, hot water extracts of the mushrooms were more potent as antimicrobial agent than either ethanol or cold water extract. It is hereby recommended that these bioactive compounds in mushrooms, ginger and garlic which show antimicrobial activities should be harnessed, patented and circulated as alternative antimicrobials to curb the increasing menace of antimicrobial resistance.

Index Terms: Antimicrobial agents, Clinical Isolates, Drug Resistance, Garlic, Ginger, Mushrooms, Nigeria.

1 INTRODUCTION

Global antimicrobial resistance is of increasing public health concern, although a number of natural and synthetic antimicrobial agents have been isolated and developed to effectively control pathogenic microorganisms [1],[2]. As a result, infectious diseases have remained a sustainable burden to human health [1],[3],[4].

This situation of rampant multiple drug resistance among human pathogenic microorganisms has necessitated a continuous search for new antimicrobial agents especially among plants with reported novel potent antimicrobial activities [5]. Also, the knowledge of feeding practices of different ethnic groups across the globe has greatly encouraged the development of research on natural products. This has led to the establishment of relationship between the chemical structure of compounds found in natural products and their biological properties as well as their importance to human health [6]. These natural products have been shown to have potential usefulness in the production of antibacterial drugs [7]. However, most herbal plants and seeds, which have indicated antimicrobial potentials, are yet to be validated of their claimed activities and possibly developing drugs from them. Mushrooms, also called puffballs are macroscopic fungi that can be found in various places such as wet environments, decayed plants and animal sites, termites nest, palm wastes, leaf litters, under shades, to mention but a few [8]. They are distinctive fruiting bodies which are either hypogeous or epigeous [9]. Mushrooms are differentiated into edible or poisonous, wild or domestic. In many countries of the world, Nigeria inclusive edible mushrooms are good for food and are used in medicine to protect against free radicals and infections [10]. Besides medicinal and nutritional use, mushroom can be used as natural dyes for fabrics. Medicinal mushrooms have been proven to be rich sources of bioactive compounds which belong to several chemical groups often polysaccharides or tripenes [2]. Sixty antimicrobial agents have been isolated from mushrooms; however, only the compounds from microscopic fungi are in circulation as antibiotics in the markets [5]. Ginger (*Zingiber officinale*) and garlic (*Allium sativum*) are spices and examples of herbal plants which in

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comparison to formulated drugs have fewer side effects [11]. Ginger, a rhizome which is white to yellow in colour and very aromatic has lots of beneficial uses both in medicine for the treatment of high blood pressure, cold-induced diseases, asthma, persistent cough and other related problems of the respiratory system [12], and in flavouring of foods [10]. Garlic is a species of onion; it possesses a characteristic hot, pungent flavor and has been known down history to be used for both culinary and medical purposes [13]. It is very easy to grow. Garlic is an antioxidant and also helpful in the treatment of common cold and cardiovascular diseases [14]. Various microorganisms have been either inhibited or killed when in contact with some of these edible mushrooms, garlic and ginger. Example includes *P. aeruginosa*, *E. coli*, *Bacillus cereus*, and *Salmonella typhi* [15],[16]. The present study, therefore, investigated the antimicrobial activities of extracts of seven edible mushrooms and two spices (ginger and garlic) against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Streptococcus pyogenes*, *Escherichia coli*, and *Candida albicans* from Federal University Teaching Hospital, Abakaliki (FETHA), Ebonyi State, Nigeria

2 MATERIALS AND METHODS

2.1 Collection and Identification of Materials

Seven species of mushrooms (*P. campestris*, *Boletus* sp, *Cortinarius* sp, *Trichaptum* sp, *Tricholoma nudum*, *Auricularia auricular-judae*, and *Flammulina* sp) were collected from different parts of Egugwu, Umuchima, and Umunaga, all in Uburu, Ohaozara Local Government Area, Ebonyi state, Nigeria. The garlic and ginger used were bought from Abakpa market, Abakaliki in Ebonyi state.

2.2 Test Organisms Used

Pure cultures of *S. pyogenes*, *P. aeruginosa*, *E. coli*, *S. aureus*, *Salmonella typhi*, were collected from the Medical Microbiology Laboratory of Federal University Teaching Hospital, Abakaliki (FETHA), Ebonyi State, Nigeria. Whereas the pure cultures of *C. albicans* were collected from Biotechnology Research and Development Centre, Ebonyi State University, Abakaliki, Nigeria.

2.3 Extraction of Mushrooms, Ginger and Garlic

The extraction was carried out in accordance with the procedure of Fasidi and Jonathan [16]. The respective mushroom, ginger and garlic samples were cut into bits and sun dried for 6 days and then pulverised into powder using manual grinder. 40g each of the pulverised mushroom, were respectively weighed and soaked in 200ml of cold water, hot water, ethanol whereas the ginger and garlic were soaked in 200ml of cold water only. Cold water preparation was allowed to stand for 2 days only with interval shaking at 30 minutes (this is to prevent mold from growing in the extraction water). The hot water was allowed to stand for 24hrs (for same reason as stated above). The ethanol and diethyl ether were allowed to stand for 7 days. Then the four preparations were filtered using Whatman filter paper no. 1. The filtrate was poured in crucibles and air dried at room temperature to recover extracts. The media used was prepared following the manufacturer's instruction as stated by Cheesbrough [17]. The turbidity of the bacterial suspension was standardized using McFarland's solution.

2.4 Chemicals

All chemicals used for various analyses were purchased from Aldrich chemicals, Poole, UK.

2.5 Antimicrobial Analysis of Mushrooms, Ginger and Garlic

The extracts from mushroom, ginger, and garlic were weighed at concentrations of 0.01g, 0.1g, 1.0g and dissolved in 2mls of distilled water. Agar well diffusion method was used for the analysis. Then the setups were allowed to stand for about 15mins before putting it into the incubator.

3 RESULTS

The results revealed that the hot water extracts of mushrooms showed highest antimicrobial activity when used against *Salmonella typhi* with zone of inhibition diameter of 25.0 mm and least activity when used against *Candida albicans* and *Pseudomonas aeruginosa* each producing 9.0 mm zone of inhibition diameter. On the other hands, the cold water extracts of mushrooms showed highest activity against *P. aeruginosa* and least activity against *Escherichia coli* with zones of inhibition diameter of 25.0 mm and 10.0 mm respectively. With the cold water extracts of the spices (ginger and garlic), no antimicrobial activity was observed against *E. coli*. However, ginger showed highest antimicrobial activity against *Staphylococcus aureus* and least activity against *P. aeruginosa* with zones of inhibition diameter of 25.0 mm and 11.0 mm respectively. Details of the results are as can be seen in the following Tables 1 and 2.

4 DISCUSSION

Herbal plants and spices are among the most commonly used antimicrobial agents in food and have been in use traditionally for thousands of years in the control of various health complications including infectious diseases [13]. In this study, antimicrobial activities of mushrooms, ginger and garlic were examined using the agar well diffusion method. The results indicated that extracts from mushroom, ginger and garlic have antimicrobial properties as also reported by Oliver [18]. It is interesting to note that the pathogenic microorganism, *Pseudomonas aeruginosa*, which is resistant to conventional synthetic antibiotics like gentamycin and tetracycline [19],[20] was found to show susceptibility to cold

Table 1: Antimicrobial Activities of Hot Water Extract of Mushrooms on Test Organisms

Conc. (mg/ml)	Zone of Inhibition Diameter (mm)									
	<i>T. nudum</i>		<i>A. auriculajudae</i>	<i>P. campestris</i>			<i>Flammulina spp.</i>		<i>Trichaptum spp.</i>	
	<i>S. aureus</i>	<i>S. typhi</i>	<i>S. aureus</i>	<i>C. albicans</i>	<i>P. aeruginosa</i>	<i>E. coli</i>	<i>S. typhi</i>	<i>C. albicans</i>	<i>S. aureus</i>	<i>P. aeruginosa</i>
0.01	10.0	15.0	-	-	10.0	-	10.0	-	10.0	-
0.1	14.0	18.0	15.0	9.0	11.0	11.0	15.0	10.0	15.0	9.0
1.0	11.0	25.0	17.0	10.0	12.0	15.0	20.0	11.0	20.0	10.0

Table 2: Antimicrobial Activities of Cold Water Extract of Mushrooms and Spices on Test Organisms

Conc. (mg/ml)	Zone of Inhibition Diameter (mm)												
	<i>P. campestris</i>	<i>Trichaptum sp.</i>			Garlic (<i>Allium sativum</i>)				Ginger (<i>Zingiber officianale</i>)				
	<i>P. aeruginosa</i>	<i>S. aureus</i>	<i>E. coli</i>	<i>E. coli</i>	<i>P. aeruginosa</i>	<i>S. typhi</i>	<i>S. pyogenes</i>	<i>S. aureus</i>	<i>E. coli</i>	<i>P. aeruginosa</i>	<i>S. aureus</i>	<i>S. typhi</i>	<i>S. pyogenes</i>
0.01	14.0	-	-	-	-	-	-	-	-	-	-	-	-
0.1	15.0	-	11.0	-	13.0	15.0	15.0	18.0	-	11.0	20.0	18.0	18.0
1.0	25.0	11.0	14.0	-	14.0	18.0	18.0	20.0	-	13.0	25.0	20.0	20.0

water extracts of mushroom. Mushrooms produce various antiviral, antifungal compounds to survive in the wild against competing or pathogenic agents [21],[22]. Also observed in this study is that there were variations in the degree of antimicrobial activities of mushrooms. This result is in agreement with the reports of Akyuz *et al.* in Turkey [1] and that of Jaggadish *et al.* [23]. The broad spectrum activity of mushrooms was also brought to light as the extracts of mushrooms showed inhibitory effects on clinical isolates used for this investigation. This suggests that the bioactive products which are contained in mushrooms are in concentrations which exude varying degrees of antimicrobial activity. Furthermore, the study revealed that the bacterial isolates (*Pseudomonas aeruginosa*, *Escherichia coli* and *Staphylococcus aureus*) showed more sensitivity to extracts from mushrooms (Table 1) than fungal isolate (*Candida albicans*). This is in consonance with the finding of Gbolagade and Fasidi [24] which explains that the extracts of mushrooms possess low antifungal properties and may not be effective in the treatment of diseases caused by fungi. Also extracts of garlic and ginger as indicated by the results (Table 2) are seen to have a wide spectrum of antimicrobial activity with varying degrees of sensitivity when used against the clinical isolates. However, the data from Table 2 revealed that extracts from the spices had antibacterial activities against all tested bacterial isolates except *E. coli*. This finding is in accordance with the findings of Belguith *et al.* [25] and Yin *et al.* [26], Bahkt *et al.* [27], Iwalokun [28], Gull *et al.* [11] and Atai *et al.* [29]. It is interesting to note from the results of this study that clinical isolates both Gram positive and Gram negative bacteria were sensitive to the extracts. But the gram positive bacteria showed more sensitivity than gram negative bacteria. This is in collaboration with the findings of Onyeagba *et al.* [30] and Desouza *et al.* [31]. The sensitivity of isolates to the mushrooms and spices extracts implies that intrinsic substance in the extracts is unknown to the microorganisms which made it impossible for them to resist. Also, when the effects of cold water and hot water extracts on the mushrooms and spices are compared with regard to the inhibition of microbial growth, the result showed that the cold water had

greater inhibiting effect than the hot water. This is in agreement with the reports of Bingelli *et al.* [32] and Jang and Hyung [22]. The results showed that the zones of inhibition exhibited on the agar plates by the mushroom and spices extracts were concentration-dependent. The higher the concentration of the extracts, the larger the zones of inhibition produced.

5 CONCLUSION

This research has further illuminated the medicinal value of some edible mushrooms and spices found in Ebonyi State Nigeria. Most of the mushrooms tested had significant antimicrobial activity. From the present study, it can be concluded that mushroom, garlic and ginger possess good quantities of compounds which have potent antimicrobial activity. Therefore, they have lots of potentials for use in the production of novel drugs and medicines, considering the lingering threat of multi-drug resistance. It is important that these bioactive compounds be identified, isolated, quantified and properly examined for its side effects and toxicity. Furthermore, clinical evaluation of spices and mushrooms through *in vivo* based research is highly recommended to achieve low cost, less side effect treatment and prevent recurrent infections.

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