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Research article

Studies on Antimicrobial Potentials of three *Ganoderma* species

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ABSTRACT: The fruit bodies of three Ganoderma species namely *G. lucidium, G. applanatum and G. australe* were collected from the decaying logs within the University of Ibadan Botanical Gardens .Invitro antagonistic effect of the ethanol, methanol and distilled water extracts of these macro fungi were tested against some disease causing microorganisms. Both crude and pure extracts of these *Ganoderma* species exhibited various degree of inhibition against the test organisms. The widest inhibitory zone (20.3mm) were obtained with the crude methanolic extract of *G. lucidium* against *Proteus mirabilis* while the highest in-vitro antifungal activity (24.3mm) was observed in the crude ethanolic extracts of *G. lucidium* against *Aspergillus niger*. The lowest zone of inhibition (2.3mm) was demonstrated with the aqueous extract *of G. australe* against *Escherichia coli* and 2.7mm with purified extract of *G. australe* against *Penicillum oxalium* . The minimum inhibitory concentration (MIC) for the ethanol extract ranged between 1.7 and 5.0mg/ml for bacteria and between 2.0 and 6.0mg/ml for fungi. The implications of these findings were discussed.

Key Word: *Ganodermaspecies,*Antimicrobial potentials,microorganisms, extraction, Botanical Gardens

INTRODUCTION

Ganoderma species are regarded as higher fungi because the carpophores are visible enough to be seen with naked eyes. Although, the real organism comprises of intercillary microscopic bodies which could not be visualized with ordinary eyes. (Zoberi, 1972; Jonathan, 2002) .They are regarded as polypores because they possessed tiny pores underside their cap which contained reproductive spores.The caps are spongy when fresh, hardening to a shiny, smooth woody structure when matured. The colour of the caps ranges from brown, to yellowish, with reddish-brown being typical of the polypore. The pore surface is cream in colour and the spores are brown. (Zoberi, 1972).

Ganoderma species belong to the division Basidiomycota, class Homobasidiomycetes, order Aphyllophorales, family Polyporaceae. (Alexopolus *et al* 1996;Wasser and Weis, 1999a). They are numbered among several species of wood degrading fungi.(Jonathan *et al* 2008). *Ganoderma* species are not listed among the group of edible mushrooms because the fruiting bodies are always thick, corky and tough and, do not have the fleshy texture characteristics of true edible fungi. (Jong and Birmingham,1991; Jonathan *et al*, 2008). Although *Ganoderma* species could not be eating directly, they have been known all over the world as highly medicinal mushrooms (Yoon *et al* 1994; Wasser and Weis, 1999b). These macrofungi have attracted great attention all over the world because of their wide range of pharmacological values.

They have been known for their antihydrogenic, antitumor, antihepatotoxic, antinocieptic, immunodulatory, cardiovascular, antibacterial and antiviral values (Chang and Buswell,1996;Chang and Mshigeni,2001). Sheena *et al* (2003), reported that the major secondary metabolites of *Ganoderma lucidum*

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are ganoderic acid,triterpenes and carcinostatic polysaccharides. These bioactive compounds have been implicated for their highantioxidant , immunoregulatory andhypoglycemic activities.Over the past three decades, scientists all over the world have isolated more than 150 triterpenes and 50 pharmacologically active polysaccharides from different *Ganoderma* species(Kim and Kim,2002;Lin and Chou,1984; Jong and Birmingham,1991)

In West Africa especially Nigeria, many health claims have been made on the effect that *Ganoderma* species have on the immune system. Herbalists usually consider *Ganoderma* as natural regulator, suppressing the immune system if it is overactive and boosting it if it is underactive(Gao and Yang, 1991;Jonathan, 2002) The local traditional doctors among the Yoruba people of south-western Nigeria have used *Ganoderma* species in the treatment of skin disorder, high blood pressure and intestinal disorder. They usually regard *Ganoderma lucidum* as immune booster especially when combined with other medicinal ingredients. Despite the important medicinal uses of *Ganoderma* in Nigeria, information on the bacteria and fungi that they specifically controlled is scantly in Literatures. Therefore, the objective of this work is to shed light on specific pathogenic microorganisms which could be inhibited by this group of medically important mushrooms.

MATERIALS AND METHODS

Mushrooms and Test microorganisms: Three *Ganoderma* species were used in this study. These were *Ganoderma lucidum, Ganoderma applanatum and Ganoderma australe* .They were found to be growing wildly within the premises of University of Ibadan Botanical Gardens, Ibadan Nigeria. The fruit bodies of *Ganoderma lucidum* were collected from *dead* trunk of *Delonix regia,Ganoderma applanatum* from decaying logs of *Magnifera indica and Ganoderma australe* frompartially buried dead root of *Terminalia ivorensis* The test microorganisms used for these studies were *Escherichia coli*, *Pseudomonas aeruginosa*, *Proteus mirabilis*, *Klebsiella pneumoniae*, *Staphylococcus aureus, Bacillus cereus, Penicillum oxalium, P. chrysogenum, Actinomyces sp, Candida albicans, Pache dermatitis, Malassezia sloffiae, Malassezia sympodialis, Aspergillus niger, Aspergillus flavus, Aspergillus tamarri* and *Fusarium oxyporum.* These *microrganisms* have already been identified and characterized

Crude extracts

The sporophores of the three *Ganoderma* species were sun dried for two weeks. Each species was milled separately to obtained fine powdered materials. These were extracted in water, methanol and ethanol to obtained pharmacologically active compounds from *Ganoderma* species. The cold extraction procedures of Jonathan and Fasidi (2005) were used.

Purification of extracts.

The extracts were purified using the procedures of Hirasawa *et al* (1999). The suspension thus obtained was centrifuged to remove the insoluble matter; the aqueous supernatant was concentrated using rotary evaporator under reduced pressure to obtain pure extracts.(Jonathan and Fasidi,2003)

Antibacterial activities

This research was aimed at screening anti-bacterial potency of the *Ganoderma* species used. The microorganisms were sub cultured into Petri dishes and incubated for 24 hrs at 37^0C for bacteria and 72hours at 28^0C for fungi to obtained actively growing cultures at exponential phase .Determination for antibacterial activities of water, methanolic and ethanolic extracts were carried out using filter paper disc and agar well diffusion methods of Stoke and Ridgway(1980). Whatman filter paper no1 were cut into sizes (7mm diameter discs).These were autoclaved at 1.02kgcm^{-2} pressure and temperature of 121^0C for 20mins and allowed to cool. The sterile discs were impregnated with the test extracts and dried at 40 ^{0}C for 1 hour. Each disc was introduced onto the Muella Hinton agar bacteria seeded plate and placed in the cold incubator at 8^0C for 10hours to enhance the diffusion of extract into the culture medium. The Petri dishes were incubated for 24hours at $35^0 \pm 2^0$C. The zones of inhibition were then observed and measured with the aid of metre rule. Each treatment was replicated thee times.

For the agar well diffusion, 7mm sterile cork borer was used to make well on sterile Muella Hinton agar. 0.25ml of the extracts was introduced into the bore agar wells using sterile dropping pipette. The extracts were allowed to diffuse before inoculating with the test organisms and incubated. After 24 hours the Inhibitory zones were measured with metre rule and recorded appropriately.

Antifungal activities.

The determination for antifungal activities of these Ganoderma species were determined using *Candida albicans,Pache dermatitis, Malassezia sloffiae, Malassezia sympodialis, Aspergillus niger, Aspergillus flavus, Aspergillus tamarri andFusarium oxyporum* respectively. Sterile Saboraud dextrose agar (SDA)were prepared using standard methods. Wells

were made on the prepared sterile culture medium (SDA) using7mm sterile cork borer. About 0.2ml of the extract was introduced into the bore wells on the agar using sterile dropping pipette. The extracts were allowed to diffuse overnight inside the refrigerator before seeded with the test fungi and incubated at $30^0 \pm 2^0$C for 3-5days. The plates were examined for any zone of inhibition which was measured in millimetres (mm).

Minimum Inhibitory Concentration (MIC)

This study aimed in finding out the lowest concentration of methanol, ethanol and distilled water extracts that will inhibit the growth of the test microorganisms.

Different concentrations varying from 1.0 to 18.0mg/ml of the extracts were prepared. The test was carried out using hole diffusion method. The highest concentrations of the extracts were first tested followed by less concentrated extracts until no inhibitory zones were observed. The lowest concentration (dilution) at which inhibitory zone was produced is regarded as the minimum inhibitory concentration. (MIC) for each extract (Jonathan and Fasidi, 2003). Each experiment was carried out in triplicates to ensure precision and the inhibitory zones were measured accordingly. The sterile distilled water without any *Ganoderma* extract served as the control.

RESULTS

Table 1 shows results of inhibitory action of three*Ganoderma* extracts on the test bacteria. It was observed that all the screened higher fungi (*G. lucidum, G. applanatum*, and *G. australe)* demonstratedvarious degrees of antibacterial activities. For ethanolic extract, the highest antibacterial activity (18.3mm) was demonstrated by *G.lucidum* purified extract against *Bacillus cereus.* The crude extract of the same fungus produced 11.0mm zone of inhibition for *B.cereus* (Table 1).

Table 1: Antibacterial Activities of Ethanolic Extracts of the Three *Ganoderma* Species

Test bacteria	Zone of inhibition (mm).Mean of the three replicates					
	Glu E^c	Glu E^p	Gap E^c	Gap E^p	Gau E^c	Gau E^p
E. coli	14.3^{c}	10.7^{e}	8.3^{d}	10.7^{d}	8.7^{c}	7.3^{cd}
P. aeruginosa	10.3^{de}	12.3^{d}	6.0^{e}	3.3^{f}	11.7^{b}	11.3^{ab}
P. mirabilis	16.0^{b}	3.3^{f}	5.7^{ef}	7.3^{e}	0	3.3^{e}
K. pneumoniae	11.7^{d}	15.0^{b}	8.7^{cd}	15.7^{a}	8.0^{cd}	9.7^{b}
S. aureus	17.7^{a}	14.3^{c}	12.3^{b}	13.7^{b}	13.3^{a}	12.3^{a}
B .cereus	11.0^{e}	18.3^{a}	15.7^{a}	10.7^{d}	8.7^{c}	8.3^{c}
Actinomyces spp	8.3^{f}	11.3^{de}	5.3^{f}	12.0^{c}	0	2.7^{ef}
Control (DW)	0	0	0	0	0	0

DW=distilled water; Glu E^c = *Ganoderma lucidium*crude extract; Glu E^p = *Ganoderma lucidium* purified extract
Gap E^c = *Ganoderma applanatum* crude extract; Gap E^p = *Ganoderma applanatum* purified extract; Gau E^c = *Ganoderma australe* crude extract; Gau E^p =*Ganoderma australe* purified extract
Values follows by the same superscript(s) along each column are not significantly different by Duncan's multiple range test.(P > 0.05).

Table 2: Antibacterial activities of methanolic extracts of the three *Ganoderma* species

Test bacteria	Zone of inhibition (mm).Mean of the three replicates					
	Glu E^c	Glu E^p	Gap E^c	Gap E^p	Gau E^c	Gau E^p
E. coli	10.3^{c}	16.7^{b}	11.0^{e}	8.3^{f}	12.7^{b}	10.3^{de}
P. aeruginosa	11.7^{bc}	14.3^{c}	15.7^{c}	15.0^{cd}	6.3^{f}	6.7^{f}
P. mirabilis	20.3^{a}	18.0^{a}	17.7^{b}	18.7^{a}	11.0^{d}	12.7^{cd}
K. pneumoniae	12.0^{b}	8.3^{f}	19.3^{a}	16.0^{b}	7.7^{e}	8.7^{e}
S. aureus	8.0^{d}	11.0^{d}	12.0^{de}	15.0^{c}	14.0^{a}	16.7^{a}
B .cereus	4.0^{ef}	9.3^{ef}	12.7^{d}	14.3^{d}	12.3^{cd}	15.3^{ab}
Actinomyces spp	3.7^{f}	8.3^{f}	12.7^{d}	14.3^{d}	12.3^{cd}	15.3^{ab}
Control (DW)	0	0	0	0	0	0

DW=distilled water; Glu E^c = *Ganoderma lucidium*crude extract; Glu E^p = *Ganoderma lucidium* purified extract
Gap E^c = *Ganoderma applanatum* crude extract; Gap E^p = *Ganoderma applanatum* purified extract; Gau E^c = *Ganoderma australe* crude extract; Gau E^p =*Ganoderma australe* purified extract

Values follows by the same superscript(s) along each column are not significantly different by Duncan's multiple range test.($P > 0.05$).

Table 3: Antibacterial activities of water extracts of the three *Ganoderma* species

Test bacteria	Zone of inhibition (mm).Mean of the three replicates					
	Glu E^c	Glu E^p	Gap E^c	Gap E^p	Gau E^c	Gau E^p
E. coli	10.3^b	7.3^d	12.3^a	5.0^{de}	2.3^d	0
P. aeruginosa	6.7^c	5.0^{ef}	5.7^e	4.3^e	0	2.7^c
P. mirabilis	4.7^d	5.7^e	10.7^a	9.0^c	9.7^a	9.0^a
K. pneumoniae	9.3^b	10.7^c	8.7^c	17.7^a	0	3.3^c
S. aureus	11.0^a	15.7^a	9.3^b	5.3^d	2.7^d	0
B .cereus	11.7^a	9.0^c	7.7^d	12.3^b	7.3^b	7.7^b
Actinomyces spp	11.0^a	13.0^b	10.0^{ab}	11.7^b	6.3^c	8.3^{ab}
Control (DW)	*0*	0	0	0	0	0

DW=distilled water; Glu E^c = *Ganoderma lucidium*crude extract; Glu E^p = *Ganoderma lucidium* purified extract
Gap E^c = *Ganoderma applanatum* crude extract; Gap E^p = *Ganoderma applanatum* purified extract; Gau E^c = *Ganoderma australe* crude extract; Gau E^p =*Ganoderma australe* purified extract
Values follows by the same superscript(s) along each column are not significantly different by Duncan's multiple range test.($P > 0.05$).

Table 4: Antifungal activities of Ethanolic extracts of the three *ganoderma* species

Fungal species						
	Glu E^c	Glu E^p	Gap E^c	Gap E^p	Gau E^c	Gau E^p
A. niger	24.3^a	13.3^e	$20.7^{a,b}$	23.3^a	14.3^a	12.3^a
A . flavus	11.7^g	10.3^g	8.3^e	6.3^h	0	0
A . tamarri	18.3^c	20.7^a	21.7^a	18.7^d	0	0
C . albicans	21.0^c	15.3^d	16.0^c	21.0^c	10.7^{cd}	8.3^c
F. oxyporum	14.7^d	12.0^{ef}	4.0^g	4.0^i	0	0
M. sloffiae	16.7^c	11.7^f	21.3^a	15.7^f	11.0^{bc}	12.3^a
M. sympodialisis	14.3^e	18.3^b	19.3^b	22.0^{abc}	9.0^d	10.7^b
P .dermatitis.	22.0^{bc}	$11.0^{f,g}$	12.0^d	10.7^g	11.3^{bc}	8.7^c
P . chrysogenum	12.3^{fg}	17.0^{bc}	16.3^c	18.3^d	0	6.7^d
P. oxalium	11.0^f	6.3^h	6.7^f	10.0^g	1.7^e	0
Control (DW)	0	0	0	0	0	0

Gap E^c = *Ganoderma applanatum* crude extract; Gap E^p = *Ganoderma applanatum* purified extract; Gau E^c = *Ganoderma australe* crude extract; Gau E^p =*Ganoderma australe* purified extract
Values follows by the same superscript(s) along each column are not significantly different by Duncan's multiple range test.

Moderate zone of inhibition were produced by both crude and purified extracts of *G.lucidum* against *Proteus mirabilis,Staphyloccoccus aureus* and *Klebsiella pneumoniae* .The lowest antagonistic effect of the ethanolic extracts was observed with *G.applanatum* against *Actinomyces spp* .

Table2 shows the antibacterial activities of crude and purified methanolic extracts against the selected bacteria.*G. lucidun*crude extract demonstrated the highest activity(20.3mm) against *K. pneumonia,* closely followed by the *G. applantatum* crude extract(19.3mm) against *P.mirabilis.*Nozone of inhibition was seen in *G.australe* against *Actinomyces spp.*

Antibacterial potential of the three *Ganodema* species using water as solvent for extraction was presented on Table 3.Generally , water extracts of all the studied mushrooms demonstrated lower values of antibacterial activities compared with methanol and ethanol. The widest zone of inhibition (17.7mm) was seen in *G. applanatum* purified aqueous extract against *K. pneuminiae* .Likewise, *G.lucidum* purified aqueous extract produced 15.7 mm zone of inhibition against *S. aureus* .

The antifungal potential of crude and purified extracts of the three Ganoderma species were presented on Table 4 .The greatest zone of inhibition (24.3mm) was produced by the crude extract of *G. lucidum* against *A.niger* . Purified extract of *G. applanatum* produced the second best zone of inhibition(23.3mm) against the same fungus .The crude extract of *G. lucidum* and purified extract of *G.applantatum* had 22.0mm inhibitory zones against *P.dermatitis* and *M.sympodialis* respectively .*Candida albicans* was also well inhibited by the crude extract of *G. lucidum*

and purified extract of *G. applanatum* with 21.0mm inhibitory zone.

Table 5: Antifungal activities of methanolic extracts of the three *Ganoderma* species

Fungal species	Glu E^c	Glu E^p	Gap E^c	Gap E^p	Gau E^c	Gau E^p
A. niger	11.7^{e}	6.3^{g}	13.7^{e}	12.7^{e}	11.7^{c}	8.0^{de}
A . flavus	21.3^{a}	17.3^{b}	15.7^{d}	9.0^{f}	12.7^{bc}	11.0^{c}
A . tamarri	5.0^{h}	10.3^{f}	8.3^{h}	6.7^{h}	0	0
C . albicans	15.3^{b}	13.3^{e}	22.3^{a}	19.7^{ab}	16.0^{a}	10.7^{c}
F. oxyporum	13.0^{d}	10.7^{f}	13.3^{f}	20.7^{a}	13.3^{b}	15.0^{a}
M. sloffiae	14.0^{cd}	21.7^{a}	16.3^{c}	17.7^{c}	11.0^{cd}	14.3^{b}
M. sympodialisis	21.0^{a}	17.0^{b}	19.0^{b}	13.3^{d}	10.3^{d}	8.3^{d}
P .dermatitis.	13.3^{d}	15.3^{cd}	6.0^{i}	19.3^{ab}	1.7^{f}	7.0^{e}
P . chrysogenum	15.3^{b}	17.7^{b}	10.7^{g}	9.3^{f}	0	0
P. oxalium	6.7^{f}	14.3^{de}	8.7^{h}	7.3^{g}	4.7^{e}	2.7^{f}
Control (DW)	0	0	0	0	0	0

Gap E^c = *Ganoderma applanatum* crude extract; Gap E^p = *Ganoderma applanatum* purified extract; Gau E^c = *Ganoderma australe* crude extract; Gau E^p =*Ganoderma australe* purified extract

Values follows by the same superscript(s) along each column are not significantly different by Duncan's multiple range test.

Table 6: Antifungal potential of water extracts of the three *Ganoderma species*.

Fungal species	Glu E^c	Glu E^p	Gap E^c	Gap E^p	Gau E^c	Gau E^p
A. niger	6.3^{h}	10.3^{g}	7.0^{g}	8.7^{e}	0	0
A . flavus	13.7^{d}	11.7^{e}	13.0^{cd}	11.0^{cd}	8.7^{d}	8.0^{f}
A . tamarri	3.3^{i}	10.0^{g}	3.7^{h}	8.0^{f}	0	0
C . albicans	18.7^{a}	22.0^{a}	15.3^{b}	12.3^{c}	11.0^{b}	12.3^{cd}
F. oxyporum	8.0^{g}	11.3^{f}	9.3^{f}	15.3^{a}	7.0^{e}	15.3^{a}
M. sloffiae	13.0^{d}	20.7^{b}	18.3^{a}	14.7^{a}	13.7^{a}	14.7^{a}
M. sympodialisis	16.3^{b}	18.7^{c}	15.7^{b}	13.7^{ab}	9.0^{cd}	13.7^{bc}
P .dermatitis.	15.3^{c}	12.7^{e}	12.7^{d}	13.0^{b}	6.7^{e}	13.0^{bc}
P . chrysogenum	8.3^{f}	14.7^{d}	11.7^{e}	12.7^{c}	9.3^{c}	12.7^{cd}
P. oxalium	10.0^{e}	11.0^{f}	9.0^{f}	10.3^{d}	0	10.3^{e}
Control (DW)	0	0	0	0	0	0

Gap E^c = *Ganoderma applanatum* crude extract; Gap E^p = *Ganoderma applanatum* purified extract; Gau E^c = *Ganoderma australe* crude extract; Gau E^p =*Ganoderma australe* purified extract

Values follows by the same superscript(s) along each column are not significantly different by Duncan's multiple range test.

Ganoderma australe extracts demonstrated low antifungal activities against *C. albicans*(Table 4).The lowest antifungal activity(1.7mm) was recorded with the crude extract of *G. australe* against *P.oxalium*.All these results were statistically significant compared with the control (distilled water)experiments(P>0.05) .

On Table 5, the results of antagonistic activities of *Ganoderma* species against some selected fungi pathogens were represented. The purified extract of G. *lucidum* had the highest activity (21.7mm) against *M. slofiae* Likewise, crude extract of this macro fungus also produced inhibition of 21.3mm against *A. tamari* and *M.sympodialis* respectively. The pure extract of *G. applanatum* produced 20.7 and 19.3mm inhibitory zones against *F.oxysporum* and *P.dermatitis* respectively .The least inhibition was observed with the *G. australe* extract against *P.oxalium* .

Table 6 shows that the best zone of inhibition (22.0mm) was produced by the purified water extract of *G. lucidum* against *C. albicans*..Likewise, purified aqueous extract of the same macro fungus produced 20.7mm inhibitory zone against *M.sloffiae* .The crude aqueous extracts of *G. lucidum* and*G. applanatum* produced the moderate inhibitory zones (18.7 and 18.3 mm respectively) against *C. albicans* and *M.sloffiae* .There values were not statistically different from each other (P>0.05).Generally, water extracts produced

lower inhibitory zones than methanolic and ethanolic extracts against all the tested fungi.

Table 7: Minimum Inhibitory Concentration (MIC) of thethree *Ganoderma* extracts against the test microoganisms

Fungal species	Glu E^c	Glu E^p	Gap E^c	Gap E^p	Gau E^c	Gau E^p
E. coli	9.0^e	4.7^f	2.3^h	5.0^g	ND	2.3^f
P. aeruginosa	5.0^g	6.7^e	ND	ND	6.0^c	4.8^c
P. mirabilis	11.3^c	ND	ND	1.8^i	ND	ND
K. pneumoniae	6.3^f	11.0^b	3.3^g	8.0^e	1.7^e	5.3^b
S. aureus	11.0^d	10.3^c	5.8^f	6.0^f	7.3^b	6.3^a
A . niger	16.8^a	7.8^d	15.3^b	15.8^a	8.0^a	6.0^a
A . tamarri	11.3^c	14.8^a	15.7^a	12.7^c	ND	ND
C . albicans	15.0^b	10.3^c	10.7^c	14.0^b	5.0^e	ND
P . dermatitis	15.3^b	6.3^e	6.8^e	6.0^f	5.7^d	ND
P . chrysogenum	5.0^g	11.0^b	10.0^d	12.3^d	ND	2.3^d
Control (DW)	ND	ND	ND	ND	ND	ND

Gap E^c = *Ganoderma applanatum* crude extract; Gap E^p = *Ganoderma applanatum* purified extract; Gau E^c = *Ganoderma australe* crude extract; Gau E^p =*Ganoderma australe* purified extract

Values follows by the same superscript(s) along each column are not significantly different by Duncan's multiple range test.

Table 7 shows the minimum inhibitory concentration. The lowest MIC (1.7mg/ml)was seen in *G.australe* crude extract against *K. pneumoniae*.This was followed closely by *G. applanatum* and *G.australe* crude extract against *E. coli* with 2.3 mg/ml

DISCUSSION

From this study, it was seen clearly that all the three medicinal mushrooms used (that is *G. lucidum, G. applanatum* and *G. australe)* demonstratedhigh level of antimicrobial activities in different proportions. These results affirm the claims of traditional herbalists in the south western Nigeria that *Ganoderma* species could be used to treat some bacterial and fungal infections of man. It was suggested by Oei (2003) that *Ganoderma* species especially *G. lucidum* could be used as feed supplement to resist microbial infections and boost immune system in human beings.

It was observed that G. *lucidum, G. applanatum and G.australe* extracts behaved differently in their antimicrobial effectiveness depending on the solvent used for extraction. This is in agreement with the findings of Jonathan and Fasidi(2003),that bioactive secondary metabolites of mushrooms extracted may be different depending on the extractive solvents used .Water was observed in this study to be poor solvent compared with methanol and ethanol. This result is in line with that that of Kawagishi *et al* (1988) that some Phytochemicals are more soluble in alcohol than in water. Ethanol was however better than methanol possibly because of its molecular configuration. The high level of effectiveness of ethanol as solvent of extraction could be linked with higher concentration of metabolites extracted. This also confirmed the suggestion of Fujita *et al* (2005) who suggested that ethanol was better than methanol, and methanol was better than water as extracting solvents. Both the crude and the purified extracts of *G. lucidium, G. applanatum* and *G. australe* showed significant antibacterial activities against *E. coli, P. aeruginosa, P. mirabilis, K. pneumonia,S. aureus, B cereus* and *Actinomyces sp.* The purified extract of *G. lucidium* has the highest zone of inhibition against *B . cereus* and lowest zone of inhibition against *Actinomyces sp.*The crude extract of *G. australe* did not show any antibacterial activity against *P. mirabilis*and *Actinomyces sp.* These observations indicated that *Ganoderma species* could be used for the invitro control of these disease causingbacteriaLikewise, both the crude and the purified extracts of *G. lucidium* and *G. applanatum* showed antifungal efficacy against *P. oxalium, P. chrysogenum, Candida albicans, Pache dermatitis, Malassezia sloffiae, Malassezia sympodialis, Aspergillus niger, Aspergillus flavus, Aspergillus tamarri and*

Fusarium oxyporum while both the crude and the purified extracts *of G. australe* were inactive against *A. niger*, *A. tamarri*, *F. oxyporum*, *P. chrysogenum*. It was observed that the *G. lucidium* was more effective against *A. niger*with the largest zone of inhibition. This observation also confirms the claim of traditional doctors of Yoruba people of the south western Nigeria that *Ganoderma lucidum* when mixed with other medicinal ingredients could be used to treat some skin diseases in man

It was observed that different concentration of the ethanol extract of the macro fungi in sterile distilled water, posed different changes in the level of inhibition. This could be inferred that the concentration of the mushroom extracts used also play a vital role in the anti-microbial potential of the higher fungi. While some mushroom extracts would inhibit the growth of some test organisms at higher concentration, the reverse is the case for some other mushroom species. Danieli (1957) asserted that at the least MIC, the extracts will still be potent because bioactive secondary metabolites are freely available to effect antagonistic actions against the test microorganisms. Therefore, higher concentration which may poison the host cells may not be needed.

Out of the three *Ganoderma* species used,*G.lucidum* was the best macrofungus that generally exhibited high antagonistic activities against most microorganisms used. This was followed by *G.applanatum* and, *G australe*was the least. The results of this study further affirm that *G. lucidum* is a unique species of *Ganoderma that* could be used in the treatment of varieties of diseases.

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