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Assessment of antistaphylococcal activity of ethanolic extract of *Lenzites quercina* (L) P. Karsten against clinical *Staphylococcus* species

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Abstract

Background: Bioactive compounds in wild medicinal mushrooms are known to contain potent antimicrobial property. Hence, there is need to assess the antimicrobial properties of these wild mushrooms in order to adequately utilize the bioactive compounds for the production of new antimicrobials and to curb the menace of antibiotic resistance. In this study, the phytochemical property and antistaphylococcal activity of a wild medicinal macrofungus, *Lenzites quercina* was assessed.

Methods: *Staphylococcus* species were isolated from clinical samples from two well patronized hospitals in Akure, Ondo State, Nigeria. The isolates were identified based on standard microbiological methods. The sensitivity pattern of the *Staphylococcus* species against some antibiotics was assessed. The inhibitory potential of ethanolic extract of *Lenzites quercina* on the growth of the clinical Staphylococcal strains was carried out using agar well diffusion method.

Results: The percentage resistance displayed by forty two (42) *Staphylococcus* species against commonly used antibiotics ranged from 25.0 to 100. Qualitative screening and quantity of some phytochemicals in *Lenzites quercina* extract are revealed as follows: flavonoids (18.6 mg/g), saponins (2.8 mg/g), tannins (11.3 mg/g), tepernoids (1.16 mg/g) and cardiac glycosides (0.84 mg/g). Inhibitory zones displayed by extract of *Lenzites quercina* against *Staphylococcus* spp ranged from 6.0 mm to 22.0 mm at 50 mg/ml. The minimum inhibitory concentration (MIC) however varies from 6.25 mg/ml to ≥50 mg/ml. Extract of *Lenzites quercina* as well as commercial antibiotics (positive control) exhibited potent antibacterial activity against *Staphylococcus* spp.

Conclusion: The phytochemicals in *Lenzites quercina* are responsible for the anti-staphylococci activity observed. These phytochemicals can be further isolated and exploited to solve the recurrent problem of antibiotic resistance peculiar to some species of Staphylococcus.

Keywords: Hospitals, MAR, Medicinal mushrooms, Phytochemicals, Antibacterial agents

Background

Staphylococcus species is a ubiquitous human pathogen; microbiota of the skin and mucous membranes of the humans, which have the ability to colonize and infect both hospitalized and healthy people with life threatening infection [1]. *S. saprophyticus, S. epidermidis and S. aureus* are responsible for wide range of infections such as skin, soft tissue infections, bacteremia, infections of the central nervous system, bone and joints skeletal muscles, respiratory and urinary tracts, and infections associated with intravascular devices; cystitis, renal, ureteral stones and Urinary tract infection [2, 3].

Staphylococci had been recognized as major microorganisms involve in nosocomial infection due to continuous cross-resistance to beta-lactam and other antibiotics like streptomycin, tetracycline, chloramphenicol, vancomycin and erythromycin [4, 5]. Multidrug resistance is now common among the pathogenic *Staphylococci* and perhaps, greatest concern because of their intrinsic

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virulence, ability to cause a diverse array of life threatening infections and capacity to adapt to different environmental conditions [6].

The production of conventional antibiotics has increased to achieve victory over the multidrug resistance of *Staphylococcus aureus* and *Staphylococcus epidermidis* but intake of inappropriate doses and incomplete dose by patients has negated the effort. This had contributed to less effective bactericidal action of antibiotics and resulted to resistant patterns observed in some pathogenic microorganisms [7]. The overall reports on the emergence of antibiotic resistance of *Staphylococcus* species had underscored the action of new antibiotics products. Hence, phytomedicine from wild macrofungi will be a great option toward treatment of infection caused by these pathogenic organisms.

Wild macrofungi have been reported as natural source of antimicrobials [8]. Therefore, the exploitation of natural products from wild macrofungi will be a useful approach to reduce the increasing trend of multiple antibiotic resistance (MAR). Bioactive compounds such as phenolics, polysaccharides, lectins, lentinan, active hexose correlated compound (AHCC) in wild medicinal mushrooms are needed to be extracted and assessed for antimicrobial effect against resistance pathogens. *Lenzites quercina* is one of the widely available macrofungi with accumulated bioactive compounds that need to be screened for antibacterial activity. This study therefore provides information on phytochemical constituents and antistaphylococcal property of ethanolic extract of *Lenzites quercina* against some resistance and pathogenic *Staphylococcus* isolated from clinical samples.

Methods

Source of *Staphylococcus* species

Staphylococcus species used in this study were isolated from clinical samples such as urine, stool, sputum, wound swab and pus from Medical Microbiology Laboratory of State Specialist Hospital and Don Bosco Hospital, Akure, Nigeria. The methicillin resistant *Staphylococcus aureus* (MRSA) and *S. aureus* (ATCC 29213) were used as control during the experiment. The control isolates were soured from Medical Microbiology Department, University College Hospital (UCH), Ibadan, Oyo State, Nigeria.

Isolation and identification of *Staphylococcus* species

The culture media; mannitol salt agar (MSA, Oxoid) and cystine lactose electrolyte deficient (CLED, Oxoid) were prepared according to manufacturer's instruction. The clinical samples were plated on solidify media and incubated at 36 °C for 24–48 hours. Colony of *Staphylococcus* spp were sub cultured by streak method on nutrient agar (NA, Lab M) and pure colony obtained were maintained on agar slants at 4 °C. A loopful of 18 h broth containing organism was transferred to a clean glass slide, Gram stained and examined under microscope. Biochemical tests such as catalase, coagulase, urease, citrate utilization, sugar fermentation and action of isolates against novobiocin (5 μg) were carried out using the methods of [9] and [10]. The results of biochemical test were interpreted to confirm the identity of organisms according to [10, 11].

Antibiotic sensitivity test

Antibiotic sensitivity pattern of isolated *Staphylococcus* species was determined by using agar disc diffusion method following the recommendation of the Clinical and Laboratory Standards Institute [12]. The inoculum was prepared from 18 h broth culture of each isolate and adjusted to a turbidity equivalent of 0.5 McFarland Standard. Inoculum size (0.1 ml) was spread on Mueller- Hinton agar and the antibiotic discs were placed at the equidistance of the plate. The antibiotics used include; erythromycin (ERY 15 μg), cotrimoxazole (COT 25 μg), chloramphenicol (CHL 30 μg), novobiocin (NOV 5 μg), gentamicin (GEN 10 μg), streptomycin (STR 30 μg), augmentin (AUG 30 μg) and ciprofloxacin (CPR 5 μg). The zones of inhibition were measured in millimeters and compared against standard given by [12] to determine resistance and susceptibility.

Mushroom collection and preparation of extract

Lenzites quercina was collected from Aba Oyo farmland, which is 1.5 km to Oba-nla, the Federal University of Technology, Akure (Lat 07° 14^{I}N Long 05° 11^{I}E). Sample of *Lenzites quercina* was collected in October, 2014. The fruiting body was morphologically identified and confirmed by molecular tools using the internal transcribed spacer (ITS) region of the rDNA. Voucher specimen was deposited in the Department of Microbiology, Federal University of Technology, Akure (FUTA). Sample of *Lenzites quercina* was dried and ground to powder with a mill machine (Retsch GmbH 5657 HAAN). Milled sample (50 g) was soaked in 1000 ml of ethanol (95 % v/v) for 72 h. The filtrate obtained through What man No.1 filter paper was concentrated using rotary evaporator (RE-52A, UNION Laboratories, England) and dried extract was kept at 4 °C for further used.

Phytochemical screening of the extract

The presence of phytochemicals such as flavonoids, alkaloids, tannins, saponins, tepernoids, anthraquinones, steroids, cardiac glycosides, and phlobatannins were assessed using the methods described by [13, 14]. The methods are briefly described as follows:

Test for flavonoids

Extract of 0.5 g was gently warm with distilled water (2 ml) and then filtered. The filtrate was filled up to 2 ml and few drops of 10 % ferric chloride solution were

added. A green-blue or violet colouration indicated the presence of a phenolic hydroxyl group.

Test for alkaloids

Extract (0.5 g) was stirred with 5 ml of 1 % v/v aqueous hydrochloric acid (HCl) for two minutes on a steam water bath. The extract was filtered and few drops of Dragendorff's reagent were added. A prominent yellow to orange colour indicate that alkaloid was present.

Test for tannins

Ethanolic extract of *Lenzites quercina* (0.5 g) was mixed with 2 ml of solution 2 % of $FeCl_3$. A blue-green or black coloration indicated the presence of phenols and tannins.

Test for saponins

This was carried out by putting the extract (1 g) in a test tube and shaken very well. After, it was heated in a water bath and observed for the formation of stable foam, which was taken as an indication for the presence of saponins.

Test for terpenoids (Salkowski's Test)

Aliquot of extract (5 ml) was mixed with 2 mL of chloroform. Then 2 mL of concentrated sulfuric acid (H_2SO_4) was added carefully and shaken gently. A reddish brown coloration of the interphase was formed to show positive results for the presence of terpenoids.

Test for anthraquinones

Borntrager's test was used for the detection of anthraquinone. The extract (0.5 g) was shaken into 10 ml of benzene, filtered and 5 ml of 10 % ammonia solution added to the filtrate. The mixture was shaken and observed for the presences of pink red or violet color in the ammonia layer which indicates the presence of free anthraquinones.

Test for steroids

Acetic anhydride (2 ml) was added to 0.5 g of the extract and filtered. Sulphuric acid (2 ml) was added to the filtrate and observed for color change from violet to blue - green, which indicates the presence of steroid.

Test for Phlobatannins

The mushroom extracts (0.2 g) were dissolved in 10 ml of distilled water each and filtered. The filtrates were.

Test for cardiac glycosides

Extract was mixed with 2 ml of glacial acetic acid containing 2 drops of 2 % $FeCl_3$. The mixture was poured into another tube containing 2 ml of concentrated sulfuric acid. A brown ring at the interphase indicates the presence of glycosides. The quantity of the screened phytochemicals was determined using the standard methods outlined by [13–15].

Antimicrobial activity of *Lenzites quercina* extract

Antimicrobial activity of extract was determined by agar well diffusion method [16]. *Staphylococcus* species from stock culture were cultivated on nutrient broth at 32 °C for 18 h. The inoculum size was adjusted by serial dilution to obtained 0.5 McFarland turbidity standards. The extract was reconstituted in 20 % v/v of dimethyl sulfoxide (DMSO). An aliquot of 0.1 ml containing organism was aseptically transferred and evenly spread onto the dried surface of sterile Mueller Hinton agar plate. A well of 6 mm were bored in the agar plate with sterile cork borer. The extract was sterilized through membrane filter (0.22 μm) and 0.1 ml was aseptically introduced into the well in the Petri dishes already inoculated with *Staphylococcus* spp. A volume of 0.1 ml of erythromycin and metronidazole were used as positive control while 20 % of DMSO as negative control. The plates were incubated at 36 °C for 24 h. The diameter of the inhibition zones were measured in millimeters. The negative control shows no zone of inhibition against tested organisms.

Determination of minimum inhibitory concentration of the extract

Agar well diffusion method of [16] was used to screen the antimicrobial effect of the extracts at different concentrations. The minimum inhibitory concentration (MIC) was carried out on the extract that had antimicrobial effect on the isolated organisms. The concentrations of the extract ranged from 6.25 mg/ml to 50 mg/ml. The test was carried out in replicates ($n = 3$). The MIC was obtained by taking the least concentration of the extract that shows inhibitory effect on the tested organisms.

Statistical analysis

Experiments were carried out in replicates and data obtained were analyzed by one way analysis of variance (ANOVA) and means were compared by Duncan multiple range test (SPSS 17.0 version). Differences were considered significant at $P \leq 0.05$

Results

Table 1 shows the antibiotic resistance pattern of Staphylococci isolated from clinical samples in two Hospitals. The percentage of antibiotic resistance for the tested *Staphylococcus* spp ranged from 25 to 100 %. The indicator organisms were mostly resistance to chloramphenicol, augmentin, gentamicin, augmentin and erythromycin. It was observed that *Staphylococcus* isolates were resistance to at least one of the antibiotics used and no isolates were susceptible to all the tested antibiotics. The result of the phytochemical contents of *Lenzites quercin*a extract (Table 2)

Table 1 Resistance percent (%) of isolated *Staphylococcus* species from different clinical samples against some antibiotics

Organisms/source	Samples	Number of isolates	Antibiotics							
			ERY	COT	CHL	NOV	GEN	STR	AUG	CPR
SASH	Urine	3	66.7	0.0	100	0.0	33.3	33.3	66.7	0.0
SASH	Sputum	2	100	0.0	100	0.0	50.0	0.0	100	0.0
SASH	Pus	4	100	50.0	75.0	0.0	0.0	50.0	75.0	50.0
SASH	Stool	1	0.0	100	100	0.0	100	0.0	100	0.0
SASH	Wound swab	6	83.3	66.7	100	0.0	50.0	33.3	50.0	33.3
SESH	Pus	3	66.7	33.3	66.7	0.0	66.7	0.0	33.3	0.0
SESH	Wound swab	3	33.3	66.7	66.7	0.0	0.0	0.0	33.3	0.0
SSSH	Urine	4	25.0	0.0	75.0	100	25	50.0	50.0	0.0
SADB	Urine	3	100	66.7	100	0.0	66.7	33.3	0.0	33.3
SADB	Sputum	2	100	50	100	0.0	100	100	100	0.0
SADB	Pus	3	66.7	33.3	66.7	0.0	33.3	33.3	100	0.0
SADB	Stool	1	0.0	100	100	0.0	100	0.0	100	0.0
SADB	Wound swab	3	100	100	33.3	0.0	33.3	100	100	33.3
SEDB	Pus	1	0.0	100	0.0	0.0	100	0.0	0.0	0.0
SEDB	Wound swab	3	100	33.3	66.7	0.0	33.3	100.	33.3	33.3
[a]MRSA		1	0.0	100	100	0.0	100	100	100	0.0
[a]*S. aureus* (ATCC 29213)		1	0.0	100	100	0.0	100	100	100	100

Values are mean of replicates ($n = 3$), 0.0: isolate(s) are susceptible to the antibiotic, [a]isolates are used as the control antibiotic codes were defined under methods
SASH Staphylococcus aureus from State Specialist Hospital, Akure,
SESH Staphylococcus epidermidis from State Specialist Hospital, Akure,
SSSH Staphylococcus saprophyticus from State Specialist Hospital, Akure,
SADB Staphylococcus aureus from Don Bosco Hospital, Akure,
SEDB Staphylococcus epidermidis from Don Bosco Hospital, Akure

revealed the presence of flavonoids, saponins, alkaloids, tannins, tepernoids and cardiac glycosides. Flavonoids have the highest value of 18.6 mg/g while tannins, saponins and tepernoids were 11.3 mg/g, 2.8 mg/g and 1.16 mg/g respectively. Low quantity of alkaloids (0.48 mg/g) and cardiac glycosides (0.84 mg/g) was observed in the ethanolic extract of *Lenzites quercin*a.

Table 3 shows the antistaphylococcal activity of ethanolic extract of *Lenzites quercina* at 50 mg/ml. Ethanolic extract of *Lenzites quercina* exhibited inhibitory action on the tested isolates of *S. aureus, S. epidermidis* and *S. saprophyticus* collected from well patronized hospitals in Akure, Nigeria. Highest zone of inhibition (22 mm) was observed for *S. epidermidis*. The extract exhibited inhibition on the twenty–seven (27) species of *Staphylococcus* at 50 mg/ml, which is 64.3 % of the indicator organisms. The control isolates; MRSA and *S. aureus* (ATCC 29213) were also susceptible to the ethanol extract of *L. quercina* at 50 mg/ml. Table 4 shows minimum inhibitory concentrations (MIC) of the extract against tested isolates of *Staphylococcus* spp. Some species of *Staphylococcus* (35.7 %) was not inhibited by the ethanolic extract of *L. quercina* at 50 mg/ml; therefore the range of minimum inhibitory concentration (MIC) of the extract against the isolates was from 6.25 mg/ml to ≥ 50 mg/ml.

Table 2 Phytochemical constituents of ethanolic extract obtained from *Lenzites quercina*

Phytochemicals	Extract of *Lenzites quercina*	Quantity of phytochemicals (mg/g)
Flavonoids	+	18.6 ± 0.02
Alkaloids	+	0.48 ± 0.0
Anthraquinones	-	0.0
Saponins	+	2.8 ± 0.0
Steroids	-	0.0
Tannins	+	11.3 ± 0.01
Phlobatannins	-	0.0
Tepernoids	+	1.6 ± 0.0
Cardiac glycosides	+	0.84 ± 0.02

Key: – = absent, + = present, values are mean ± standard deviation

Discussion

Staphylococci had been recognized as one of the major pathogens of hospital acquired infections due to their resistance to commercial antibiotics. The isolated species of *Staphylococcus* show resistance against some tested antibiotics (Table 1). The multiple antibiotic resistances posed by species of *Staphylococcus* against most of the antibiotics had been attributed to the acquisition of

Table 3 Zones of inhibition (mm) of ethanolic extract of *Lenzites quercina* against tested species of *Staphylococcus* at 50 mg/ml

Clinical samples	Isolates	Extract	Erythromycin (20 mg/ml)	Metronidazole (20 mg/ml)
Urine	SASH-1	$8.0^{a} \pm 0.0$	$14.0^{b} \pm 0.0$	$9.3^{a} \pm 0.1$
Urine	SASH-2	$12.0^{b} \pm 0.0$	$10.0^{a} \pm 0.0$	$10.0^{a} \pm 0.1$
Urine	SASH-3	0.0^{a}	$15.0^{c} \pm 0.0$	$11.0^{b} \pm 0.0$
Sputum	SASH-4	0.0^{a}	$10.0^{b} \pm 0.0$	0.0^{a}
Sputum	SASH-5	0.0^{a}	$8.3^{b} \pm 0.1$	0.0^{a}
Pus	SASH-6	$13.0^{a} \pm 0.0$	$14.0^{a} \pm .0$	$24.0^{b} \pm 0.2$
Pus	SASH-7	$15.0^{b} \pm 0.0$	$13.0^{a} \pm 0.1$	$18.0^{c} \pm 0.0$
Pus	SASH-8	$11.0^{a} \pm 0.1$	$10.0^{a} \pm 0.1$	$13.0^{b} \pm 0.0$
Pus	SASH-9	$14.0^{a} \pm 0.0$	$20.0^{c} \pm 0.0$	$17.0^{b} \pm 0.0$
Stool	SASH-10	0.0^{a}	$18.0^{c} \pm 0.0$	$14.2^{b} \pm 0.1$
Wound swab	SASH-11	$10.0^{b} \pm 0.0$	$8.0^{a} \pm 0.1$	$17.1^{c} \pm 0.2$
Wound swab	SASH-12	$18.0^{c} \pm 0.0$	0.0^{a}	$15.2^{b} \pm 0.0$
Wound swab	SASH-13	$11.0^{a} \pm 0.3$	$17.0^{b} \pm 0.0$	$17.0^{b} \pm 0.6$
Wound swab	SASH-14	$20.0^{c} \pm 0.0$	0.0^{a}	$9.0^{b} \pm 0.0$
Wound swab	SASH-15	0.0^{a}	0.0^{a}	$18.0^{b} \pm 0.0$
Wound swab	SASH-16	$14.0^{b} \pm 0.2$	$21.0^{c} \pm 0.1$	0.0^{a}
Urine	SADB-17	$7.0^{b} \pm 0.02$	$10.0^{c} \pm 0.1$	0.0^{a}
Urine	SABD-18	$10.0^{b} \pm 0.0$	0.0^{a}	$15.0^{c} \pm 0.0$
Urine	SADB-19	$7.0^{a} \pm 0.0$	$12.0^{b} \pm 0.0$	$23.0^{c} \pm 0.0$
Sputum	SADB-20	0.0^{a}	10.3 ± 0.13	0.0^{a}
Sputum	SADB-21	0.0^{a}	$12.0^{b} \pm 0.0$	0.0^{a}
Pus	SADB-22	$17.0^{c} \pm 0.0$	$10.0^{b} \pm 0.0$	0.0^{a}
Pus	SADB-23	$12.0^{a} \pm 0.0$	$13.1^{a} \pm 0.3$	$19.0^{c} \pm 0.0$
Pus	SADB-24	$16.0^{a} \pm 0.0$	$22.0^{b} \pm 0.1$	$20.0^{c} \pm 0.1$
Stool	SABD-25	0.0^{a}	$15.1^{b} \pm 0.1$	$21.0^{c} \pm 0.1$
Wound swab	SADB-26	0.0^{a}	0.0^{a}	$18.0^{b} \pm 0.0$
Wound swab	SADB-27	$6.0^{a} \pm 0.0$	$18.0^{c} \pm 0.0$	$16.1^{b} \pm 0.3$
Wound swab	SADB-28	0.0^{a}	$15.0^{b} \pm 0.0$	$14.2^{b} \pm 0.1$
Pus	SEDB-29	$8.0^{b} \pm 0.0$	0.0^{a}	$20.0^{c} \pm 0.0$
Wound swab	SEDB-30	0.0^{a}	0.0^{a}	$18.0^{b} \pm 0.0$
Wound swab	SEDB-31	$13.0^{b} \pm 0.0$	0.0^{a}	$16.0^{c} \pm 0.0$
Wound swab	SEDB-32	0.0^{a}	$17.0^{c} \pm 0.0$	$9.0^{b} \pm 0.0$
Pus	SESH-33	0.0^{a}	0.0^{a}	$11.0^{b} \pm 0.0$
Pus	SESH-34	$10.0^{b} \pm 0.0$	$8.0^{a} \pm 0.0$	$13.0^{c} \pm 0.0$
Pus	SESH-35	$22.0^{c} \pm 0.0$	0.0^{a}	$10.3^{b} \pm 0.1$
Wound swab	SESH-36	$18.0^{c} \pm 0.3$	$14.3^{b} \pm 0.2$	0.0^{a}
Wound swab	SESH-37	$14.0^{b} \pm 0.0$	0.0^{a}	$15.0^{b} \pm 0.0$
Wound swab	SESH-38	0.0^{a}	$9.0^{b} \pm 0.0$	0.0^{a}
Urine	SSSH-39	$10.0^{b} \pm 0.0$	$11.0^{b} \pm 0.0$	0.0^{a}
Urine	SSSH-40	$16.1^{b} \pm 0.3$	0.0^{a}	$16.0^{b} \pm 0.0$
Urine	SSSH-41	$8.0^{b} \pm 0.0$	$9.0^{b} \pm 0.0$	0.0^{a}
Urine	SSSH-42	0.0^{a}	$11.0^{c} \pm 0.0$	$7.0^{b} \pm 0.0$
	*MRSA	$13.3^{c} \pm 0.2$	$11.0^{b} \pm 0.0$	$8.3^{a} \pm 0.2$
	S. aureus (ATCC 29213)	$14.3^{ab} \pm 0.1$	$15.0^{b} \pm 0.0$	$12.7^{a} \pm 0.8$

Values are mean ± sd of replicates ($n = 3$), means with different letters within a row are significantly different by Duncan ($P < 0.05$), 0.0 = no zone of inhibition observed, *isolates are used as the control

SASH1-16 Staphylococcus aureus from State Specialist Hospital, Akure,
SADB17-28 Staphylococcus aureus from Don Bosco Hospital, Akure,
SEDB29-32 Staphylococcus epidermidis from Don Bosco Hospital, Akure,
SESH33-38 Staphylococcus epidermidis from State Specialist Hospital, Akure,
SSSH39-42 Staphylococcus saprophyticus from State Specialist Hospital, Akure

antibiotic resistance genes [17, 18]. This has been a contentious issue over the last three decades and therefore requires prompt solution. The attention of researchers has been focused on alternative antimicrobial agents that are cheaper, safer and more efficacious. Macrofungi harbour a large number of biologically active molecules, which have

Table 4 Minimum Inhibitory Concentration (MIC) of *Lenzites quercina* extract against tested organisms

Tested isolates	MIC mg/ml	Tested isolates	MIC mg/ml
SASH-1	25.0	SADB-22	25.0
SASH-2	25.0	SADB-23	6.25
SASH-3	>50	SADB-24	6.25
SASH-4	>50	SABD-25	>50
SASH-5	>50	SADB-26	>50
SASH-6	50.0	SADB-27	12.5
SASH-7	12.5	SADB-28	>50
SASH-8	6.25	SEDB-29	12.5
SASH-9	50.0	SEDB-30	>50
SASH-10	>50	SEDB-31	12.5
SASH-11	6.25	SEDB-32	>50
SASH-12	12.5	SESH-33	>50
SASH-13	25.0	SESH-34	12.5
SASH-14	6.25	SESH-35	25.0
SASH-15	>50	SESH-36	6.25
SASH-16	25.0	SESH-37	12.5
SADB-17	12.5	SESH-38	>50
SABD-18	50.0	SSSH-39	12.5
SADB-19	25.0	SSSH-40	6.25
SADB-20	>50	SSSH-41	12.5
SADB-21	>50	SSSH-42	>50
[a]MRSA	50	[a]*S. aureus* (ATCC 29213)	12.5

Values are mean of replicates ($n = 3$), [a]isolates are used as the control

SASH 1–16 Staphylococcus aureus from State Specialist Hospital, Akure,
SADB 17–28 Staphylococcus aureus from Don Bosco Hospital, Akure,
SEDB 29–32 Staphylococcus epidermidis from Don Bosco Hospital, Akure,
SESH33-38 Staphylococcus epidermidis from State Specialist Hospital, Akure,
SSSH39-42 Staphylococcus saprophyticus from State Specialist Hospital, Akure

tremendous therapeutic potentials in curing various ailments; by enhancing the activity of different types of immune cells, destroying both virus-infected and cancerous cells [19]. The quality and quantity of phytochemical obtained in ethanolic extract of *Lenzites quercina* (Table 3) had been reported in other medicinal mushrooms. These phytochemicals are responsible for antibacterial and antifungal properties of the mushrooms [20] and [21]. Although, the quantity of phytochemicals varies, which could be influence by climate, season, location, substrate, solvent or extraction methods adopted. The phytochemicals in medicinal mushrooms may therefore be used as an alternative phytotherapeutic agent to promote human health.

Ethanolic extract of *Lenzites quercina* displayed inhibitory effect against *S. saprophyticus, S. epidermidis and S. aureus* isolated from clinical samples in two well patronized hospitals in Akure, Nigeria. The zones of inhibition obtained for the extract ranged from 6 mm to 22 mm while commercial antibiotics have zone of inhibition of 8 mm to 24 mm. The finding of Ameri et al. [22] had reported inhibitory zones (17–33) mm displayed by sesquiterpene obtained from selected *Ganoderma* species against different isolates of Methicillin resistant *Staphylococcus aureus* (MRSA). Suay et al. [23] had earlier screened some Spanish basidiomycetes for antimicrobial agents and their result indicated that different taxonomic group of fungi are of potential therapeutic interest. Similarly, Mustafa and Fatma [24] detected 14 mushroom species with significant antimicrobial activity against some target microorganisms such as *Staphylococcus aureus*, *Enterococcus feacalis*, *Streptococcus* spp and *Salmonella typhi*. Ethanolic extract of *Lenzites quercina* and commercial antibiotics widely exhibited inhibitory action (Table 2) against the tested pathogenic organisms. This may be as a result of *Lenzites quercina* extract showing varying degree of affinity to different target sites within bacterial cells. Cell wall of Gram positive bacteria is less complicated, it freely allows hydrophobic molecules to flow in [25], therefore the presence of phenolic compounds and other phytochemicals in the extract can easily penetrate the cell wall, interfere with cytoplasm, inhibits the production of enzymes and proteins synthesis. This present study thus supports the claims of [26] and [27]. These researchers associated the antimicrobial, anticancer, anti-inflammatory, antioxidant activities of wild macrofungi to diverse secondary metabolites. The metabolites are needed to be identified and isolated for the development of prolific and new pharmaceutical compounds.

Conclusions

The study revealed that ethanolic extract of *Lenzites quercina* contains bioactive compounds that are efficacious against clinically isolated *Staphylococcus aureus*, a notorious pathogenic agent that has developed resistance against commonly used antibiotic agents. Hence, the bioactive compounds from *Lenzites quercina* may be a good alternative to commercially available antibiotics that are becoming less effective on *Staphylococcus aureus*. The isolation and identification of the specific compound responsible for the antistaphylococcus effect of *Lenzites quercina* extract is therefore the next research focus.

Competing interests

Zeniyat Abubakar, Olusola Clement Ogidi and Victor Olusegun Oyetayo declare that they have no competing interests.

Authors' contribution

OVO and OCO designed the research study, OCO and ZA isolated the studied organisms and carried out the laboratory work. OCO analyzed the data and drafted the manuscript. OVO supervised the laboratory work and corrected the manuscript. All authors read and approved the final manuscript.

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