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Phytochemical and Antimicrobial studies on Tuber of *Gloriosa superba* L

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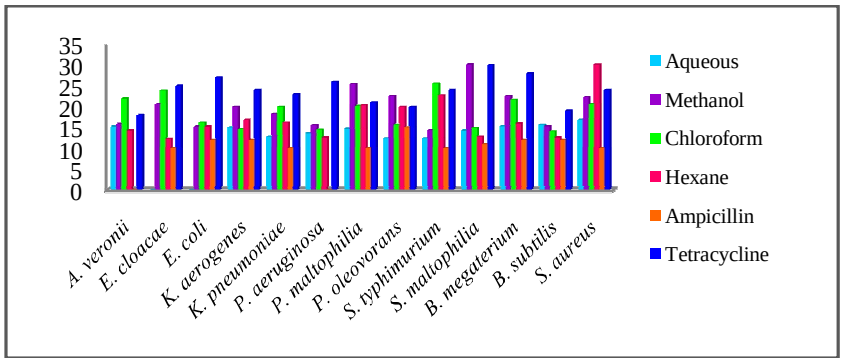
ABSTRACT

Gloriosa superba L family Colchicaceae, malabar glory lily in English. It is a medicinal important herbaceous, perennial climbing plant V-shaped rhizomes (tuber) that are white when young, and becoming brown with age found all over India. The plant usually used in traditional systems of medicine for the practice of several human diseases like cancer, gout, piles, scrofula and act as antipyretic, anti-abortive and purgative. But, it also initiated disorders and mortalities to humans and animals due to purposeful and accidental poisoning. The plant presently cultivated all over the world as an ornamental plant and medicinal herb. Alkaloid, glycosides, phenol, saponin, steroids, and tannin are present in the methanol extract of G. superba. The efficiency of antimicrobial activity from the aqueous and methanol, chloroform, hexane extracts of tuber was screened by agar diffusion method against gram-negative bacteria. The cold aqueous extract showed maximum inhibition zone of 16.8±0.2 mm in100% and13.4±0.4 mm in 50% concentrations against S. aureus. The methanol extract showed better inhibition zone of 30.2±0.1 mm in 2.5 mg mL^{-1} against S. maltophilia; 25.5±1.0 mm in 20 mg mL^{-1} against P. maltophilia and 22.6±1.4 mm in 10 mg mL^{-1} against B. megaterium. The antimicrobial effect of crude tuber extracts is better than standard antibiotic drugs ampicillin (10 mcg) and tetracycline (30 mcg). The ampicillin has shown low inhibit zone against all the organism and did not show any inhibitory effect against A. veronii and P. aeruginosa, but the Tetracycline showed a better inhibitory effect against both gram-positive and negative organism.

Graphical Abstract



Keywords: *Gloriosa superba*, Antimicrobial activity, Phytochemicals.

INTRODUCTION

Gloriosa superba L. belongs to the family Colchicaceae. Syn. *Eugonesuperba (*L.*)* Salisb., *Gloriosa angulata* Schumach., *Gloriosa cirrhifolia* Stokes, *Gloriosadoniana* Schult. and Schult.f., *Gloriosa luteaAuct.*, *Gloriosa nepalensis* G.Don, *Gloriosa rockefelleriana* Stehlé and M.Stehl., *Gloriosa rothschildiana* O'Brien, *Gloriosa superba* var. *angustifolia* Baker, *Gloriosa verschuurii* Hoog, *Gloriosa superba* f. *doniana* (Schult. and Schult.f.) T.Durand and Schinz, *Gloriosa superba* f. *grandiflora* (Hook.) Kuntze, *Methonicadoniana (*Schult. and Schult.f.*)* Kunt and *Methonica gloriosa* Salisb. Locally known as KannuvalliKodi, KalaippaikKizhangu, Sengantha malar in Tamil, Agnisikha, Agnimukhi, Ailni, Garbhaghatini, Langalika in Sanskrit, Climbing lily, Glory Lily, Malabar glory lily in English. *Gloriosa superba* occurs in grassland, semi-shade, coastal dunes, coastal woodlands, and forest in the Botswana, Eastern Cape, India, KwaZulu-Natal, Limpopo, Mpumalanga, Namibia, southeastern Asia Swaziland, tropical Africa and Zimbabwe.

It is a medicinal important herbaceous, perennial climbing plant having L or V-shaped/finger-like rhizomes (tuber) that are white when young, and becoming brown with age found all over India. The plant usually used in several traditional systems of medicine for the practice of several human diseases like cancer, gout, piles [**1**] and act as antipyretic, anti-abortive and purgative [**2**]. But, it also initiated disorders and mortalities to humans and animals due to purposeful and accidental poisoning. The plant presently cultivated all over the world as an ornamental plant and medicinal herb [**3**]. Habitually the tubers and leaves of the *G. superba* are generally used in the treatment of abdominal pain, anthelmintic, infertility, inflammation, leprosy, skin infections, piles, ulcers, [**4, 5**]. In Folklore it is being used to killing lice in the hair, pimples, and treat baldness and also as a sedative [**6**].

Nowadays, *Gloriosa superba* usually cultivated for commercial purposes. It contains the alkaloid colchicine [**7**], which used in the manufacturing of drugs. Colchicine has efficiently used in the treatment of many diseases such as abortifacient, acute gout, an antidote to snake bite, infertility, intestinal worms, laxative, skin parasites, skin problems, spines and treatment of cancer. It has also established for the treatment of arthritis, cholera, chronic ulcers, colic and kidney problems [**8**]. Colchicine is commonly used as an investigational tool for cell division in research, as it can stop mitosis and induce polyploidy. Phytochemically, in addition to colchicine also contains other compounds such as glycoside, gloriosine, long chain fatty acids, flavonoids, tannins, alkaloids, 3-O-demethylcolchicine-3-O-α-D-glucopyranoside,1,2-didemethyl colchicine, Glucoside, β and γ Lumicolichicines, 2,3-didemethyl colchicine, β sitosterol, glucoside, luteolin, N-formyl deacetyl colchicines, superbine, colchicocide, 2-hydroxy-6-methoxy benzoic and salicylic acid [**9**]. The present study attempts to evaluate the antimicrobial efficiency of the tuber of *Gloriosa superba* L.

MATERIALS AND METHODS

The tubers of *Gloriosa superba* collected from a wild area of Virudhunagar District, Tamil Nadu, India.

Successive extractive value: The tubers cut into small pieces then to dry in the shade. The shade dry tuber of *G. superba* was successive extraction with hexane, chloroform, and methanol in the order of increasing polarity of the solvent using Soxhlet apparatus [**10**].

Qualitative Analysis of Phytochemical Constituents: The methanol extracts tested for preliminary phytochemical screening [**11-13**] and the observations recorded.

Determination of Antimicrobial Activity: The antibacterial activity of aqueous, alcohol, chloroform and hexane extracts of tuber of *G. superba* were determined by agar well diffusion method [**14**]. The plant material of 200 g was weighed, chopped and divided into two portions. Each portion was crushed by grinding in a mortar and transferred to a suitable glass bottle, and a 50 mL of distilled

water was added. One glass bottle with extract was boiled (100ºC) for 20 min, and the second was mechanically shaken (200 rpm) in a cold condition for two hours. The extracts were filtered off using cheesecloth, followed by 0.45µ filter paper and transferred into a sterile closed container. The crude extract was considered as 100 % extract. By adding sterile distilled water, 50 % of the extract was prepared [**15**]. The various concentrations of 20, 10, 5 and 2.5 mg mL^{-1} solvent extracts of hexane, chloroform, and methanol of the tuber of *G. superba* were prepared for antimicrobial activity.

Test Bacteria: Thirteen human pathogenic bacterial strains *Aeromonas veronii, Bacillus megaterium, Bacillus subtilis, Enterobacter cloacae, Escherichia coli, Klebsiella aerogenes, Klebsiella pneumonia, Pseudomonas aeruginosa, Pseudomonas maltophilia, Pseudomonas oleovorans, Salmonella typhimurium, Staphylococcus aureus, Stenotrophomonas maltophila*were obtained from the National Collection of Industrial Microorganisms, National Chemical Laboratory, Pune, and used for antimicrobial studies. The strains were kept at 4°C on agar slant and subcultured at 37°C for 24 h in nutrient agar for bacteria before in *vitro* susceptibility tests.

Agar well diffusion method [**14**] was adopted to determine the antimicrobial activity. Nutrient agar (NA) plates were swabbed (Sterile cotton swabs) with 8 hours old- broth culture of respective bacteria. Two wells (8 mm diameter) were made in each of these plates using sterile cork borer, and about 0.3 mL of 100 % and 50 % aqueous extract and different concentration of plant solvent extracts were added using sterilized dropping pipettes into the wells and allowed to diffuse at room temperature for 2 h. The Petri plates were incubated for 18-24 h at 37ºC for bacterial pathogens, and individual proper control of solvent plant extracts also maintained. The diameter of the inhibition zones recorded. Triplicates were maintained, and the experiment repeated thrice, and the average values recorded for antimicrobial activity.

Statistical analysis: All experiments carried out thrice, and the data values recorded. The data were processed statistically using standard deviation and also calculated standard error. The processed data were tabulated in tables and represented in the form of a bar diagram of the graph.

RESULTS AND DISCUSSION

Preliminary Phytochemical Analysis: The methanol and chloroform extract shown the presence of secondary metabolites such as steroids, alkaloids, glycosides, terpenoids, flavonoids and steroid. Starch, saponin, phenol, and tannin are present only in methanol extracts. The terpenoid, flavone, and steroid are present in hexane extracts. These values can be used for fixing pharmacopeias standards along with other pharmacognostical and microbial parameters.

The preliminary phytochemical characters can be helpful as diagnostic tools in the exact identification of plants. The adulterants and substituents, if any in plant material can also be identified easily by these methods. The quantity of the methanol extract (6.7%) is higher than that of the chloroform (3.2 %) and hexane extracts (0.7%) of the tuber of *G. superba*. The color, appearance, and solubility of extracts indicate the quantity and nature of constituents in the extract [**16**].

Antimicrobial activity: The aqueous and methanol, chloroform and hexane extracts from an ethnomedicines *G. superba* screened for their antimicrobial efficiency. The crude extracts screened against gram-negative bacteria of *A. veronii, E. cloacae, E. coli, K. aerogenes, K. pneumonia, P. aeruginosa, P. maltophilia, P. oleovorans, S. typhimurium, S. maltophilia* and gram-positive bacteria of *B. megaterium, B. subtilis, S. aureus.* Among the different solvent extracts, the antimicrobial activity was high in the chloroform extract followed by methanol, hexane extracts. The antimicrobial activity was much less in the cold aqueous extract than boiled extract. The efficacy of antimicrobial activity depends on the nature and volume of active principles present in the tested extracts.

The aqueous extract showed activity in both gram-positive and gram-negative bacteria except *E. cloacae* , *E. coli* and *S. typhimurium*. The methanol, chloroform, and hexane extracts inhibited both gram-negative and gram-positive bacteria.

Antimicrobial activity of aqueous extract: The cold aqueous extract showed maximum inhibition zone of 16.8±0.2 mm in100% and 13.4±0.4 mm in50% concentrations against *S. aureus*. Minimum inhibition zone of 10 mm in100% and 9.7 mm in 50% concentration were observed in *P. maltophilia* (Table 1, Figure 1).The boiled aqueous extract showed maximum inhibition zone of 15.0±0. 7 mm in 100% and 12.2±0.6 mm in 50 % concentrations against *K. aerogenes*. Minimum inhibition zone of 12.4±0.7 mm in 100 % and 9.8±0.5 mm in 50 % concentration observed in P. *oleovorans* (Table 1, Figure 1).

Table 1. Antimicrobial activity of aqueous extract of *G. superba* tuber

Organism	NCIM Acc. No	Boiled Aqueous		Cold Aqueous	
		100%	50%	100%	50%
A. veronii	5621	12.0±0.2	11.6±0.4	15.2±0.8	10.0±0.7*
E. cloacae	2164	-	-	-	-
E. coli	2068	-	-	-	-
K. aerogenes	2239	15.0±0. 7	12.2±0.6	13.7±0.4	10.4±0.8
K. pneumoniae	2707	12.8±0.8	-	10.0±0.2	10.6±0.2
P. aeruginosa	3037	12.4±0. 6	10.0±0.8	13.6±0.6	10.5±0.8
P. maltophilia	2866	14.7±0.6	10.7±0.3	10.5±0.7	9.7±0.6
P. oleovorans	2867	12.4±0.7	9.8±0.5	11.9±0.1	10.2±0.6
S. typhimurium	2501	-	-	12.4±0.4	10.0±0.6
S. maltophilia	5625	14.3± 0.2	10.3±0.2	12.0±0.5	10.3±0.6
B. megaterium	2052	15.3±0.4	12.7±0.6	13.6±0.7	10.5±0.2
B. subtilis	2920	12.9±0.4	10.4±0.4	15.7±0.8	12.0±0.3
S. aureus	2079	12.0±0.6	10.2±0.9	16.8±0.2	13.4±0.4

* Zone of inhibition in mm

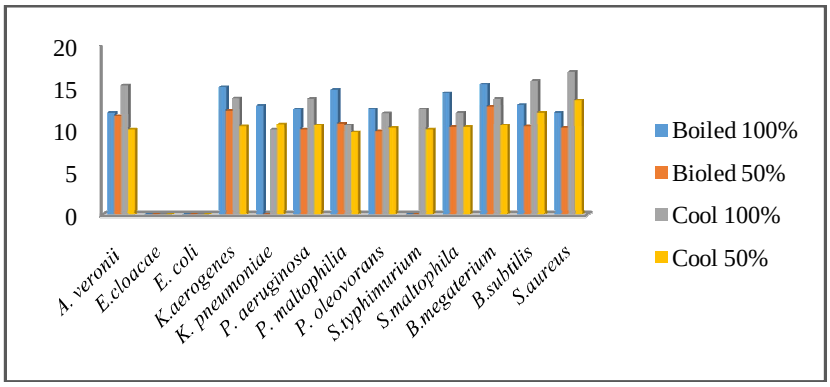


Figure 1. Antimicrobial activity of aqueous extract of *G. superba* tuber.

The antimicrobial activity of the cold aqueous extract showed better antimicrobial activity in gram-positive bacteria than negative bacteria. But, the boiled aqueous expressed the better activity against gram-negative bacteria than positive bacteria. This study revealed that the cold extract having some compound to involved the microbial activity similarly at the time of boiled the few compounds might be deactivated in the boiled extract.

Antimicrobial activity of methanol extract: The antibacterial activity of methanol extract (Table 2) showed better inhibition zone of 30.2±0.1 mm in 2.5 mg mL^{-1} against *S. maltophilia*; 25.5±1.0 mm in 20 mg mL^{-1} against *P. maltophilia*; 25.4±0.6 mm in 5 mg mL^{-1} against *S. maltophilia* and 22.6±1.4 mm in 10 mg mL^{-1} against *B. megaterium* (Figure 2). The minimum inhibitory zone (10.1±0.80) was seen in 2.5 mg mL^{-1} concentrations against *A. veronii*, 10.6±0.9 mm in 5 mg L^{-1} against *B. subtilis*,

10.6±0.9 mm in 20 mg mL^{-1} and 13.7±1.1 mm in 10 mg mL^{-1} against *A. veronii* and E. *cloacae*, and minimum inhibitory zone (10 mm) was recorded in 2.5 mg mL^{-1} concentration against *P. aeruginosa* and *S. aureus* (Table 2, Figure 2). In this study the high concentration of 20 mg mL^{-1} showed 15.3±0.4 mm but the low concentration (2.5 mg mL^{-1}) expressed the better activity (30.2±0.1) against *S. maltophilia*, because the active principle is least active when it present in higher concentration of crude extract at the same time it is active only the crude extracts may be diluted.

Table 2. Antimicrobial activity of methanol extract of *G. superba* tuber

Organism	NCIM Acc. No	Methanol extract mg mL^{-1}			
		20	10	5	2.5*
A. veronii	5621	10.6±0.9	13.7± 1.1	15.9±0.9	10.1±0.8
E. cloacae	2164	20.6±0.4	15.4±0.8	15.1±0.2	12.5± 0.7
E. coli	2068	12.7±0.9	14.8±0.4	15.2±1.4	10.9± 0.8
K. aerogenes	2239	20.0±0.7	16.4±0.6	16.0±0.2	15.3±0.1
K. pneumoniae	2707	18.3±0.8	15.8±0.6	15.5±0.9	14.3±0.8
P. aeruginosa	3037	12.5±0.9	15.5±0.8	14.3±1.0	12.8±1.3
P. maltophilia	2866	25.5±1.0	20.5 ±0.7	16.0 ±0.4	15.4±0.2
P. oleovorans	2867	18.5±0.7	18.9±0.6	22.6±0.4	20.4±0.6
S. typhimurium	2501	12.5±0.6	14.4±0.9	14.0±0.8	12.1± 0.5
S. maltophilia	5625	15.3±0.4	15.1±0.6	25.4±0.6	30.2±0.1
B. megaterium	2052	20.2±0.8	22.6±1.4	16.2 ±0.8	16.1±0.9
B. subtilis	2920	15.3±0.7	14.1±0.7	10.7±0.9	10.3±0.8
S. aureus	2079	18.2±0.7	20.4±0.9	22.3± 0.5	14.0±0.2

* Zone of inhibition in mm

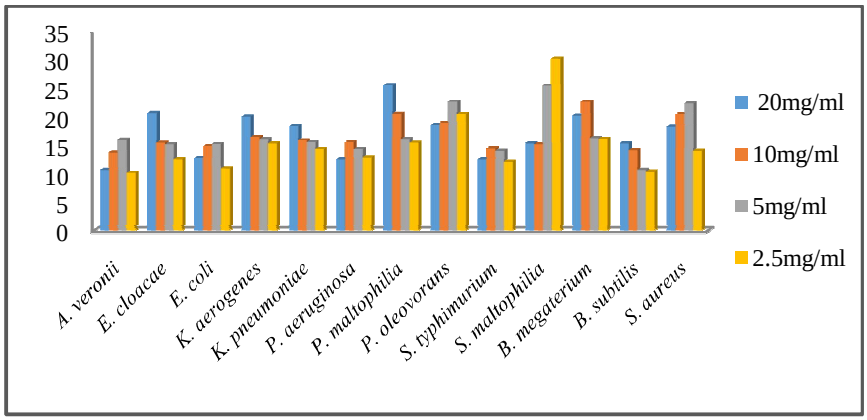


Figure 2. Antimicrobial activity of methanol extract of G. *superba* tuber.

Antimicrobial activity of chloroform extract: The antibacterial activity of chloroform extract (Table 3) showed better inhibition zone of 25.6±0.1 mm in 10 mg mL^{-1} against *S. typhimurium*; 23.9± 0.7 mm in 20 mg mL^{-1} against *E. cloacae*; 20.8±0.9 mm in 5 mg mL^{-1} and 16.3±0.2 mm in 2.5 mg mL^{-1} against *S. typhimurium* (Table 3, Figure 3).

The low inhibitory zone 10.0±0.7 showed in 2.5 mg mL^{-1} concentrations against *S. maltophilia*, 12.2±0.5 mm in 5 mg L^{-1} against *P. oleovorans*, 12.5±0.7 mm in 10 mg mL^{-1} against *P. aeruginosa*; 12.3± 0.5 mm in 20 mg mL^{-1} against *K. aerogenes*. The antimicrobial activity of the chloroform extracts expresses better activity in low concentration compare to high concentration against *S. typhimurium, S. aureus, and K. aerogenes*. The dilution factor mainly contributes to the efficiency of antimicrobial activity in chloroform extract.

Antimicrobial activity of hexane extract: The antibacterial activity of hexane extract showed better inhibition zone of 30.1±0.8 mm in 20 mg mL^{-1}, 25.3±0.2 mm in 10 mg mL^{-1} and 22.6±1.0 mm in 5 mg mL^{-1} against *S. aureus*; 18.0 ± 0.4 mm in 2.5 mg mL^{-1} against *S. typhimurium* (Table 4, Figure 4).

Table 3. Antimicrobial activity of chloroform extract of *G. superba* tuber

Organism	NCIM Acc. No	Chloroform extract mg mL^{-1}			
		20	10	5	2.5
A. veronii	5621	22.0 ±0.6	20.9±1.4	18.9±1.7	15.6±1.1*
E. cloacae	2164	23.9± 0.7	22.6±1.3	16.2±0.9	14.5 ±0.2
E. coli	2068	16.2±0.3	15.6± 0.7	14.0±0.1	10.2 ±0.6
K. aerogenes	2239	12.3± 0.5	13.3± 0.9	14.6±0.4	10.7±0.6
K. pneumoniae	2707	20.9±0.6	16.4±0.6	15.4±0.9	12.6 ±0.9
P. aeruginosa	3037	14.5±0.9	12.5±0.7	12.4±0.8	10.3 ±0.4
P. maltophilia	2866	20.2±0.8	18.6±0.3	18.2±0.6	15.1±0.7
P. oleovorans	2867	15.7±0.3	12.8±0.8	12.2±0.5	10.8±0.2
S. typhimurium	2501	22.5±0.3	25.6±0.1	20.8±0.9	16.3±0.2
S. maltophilia	5625	14.9±0.6	14.1±0.2	12.4±0.1	10.0±0.7
B. megaterium	2052	21.7 ±1.2	20.6± 0.5	17.6±0.9	15.6 ±0.7
B. subtilis	2920	14.1±0.9	12.6±0.9	12.4±0.8	10.9 ±0.9
S. aureus	2079	16.9±0.5	20.6±0.7	15.9±0.3	15.5±0.8

* Zone of inhibition in mm

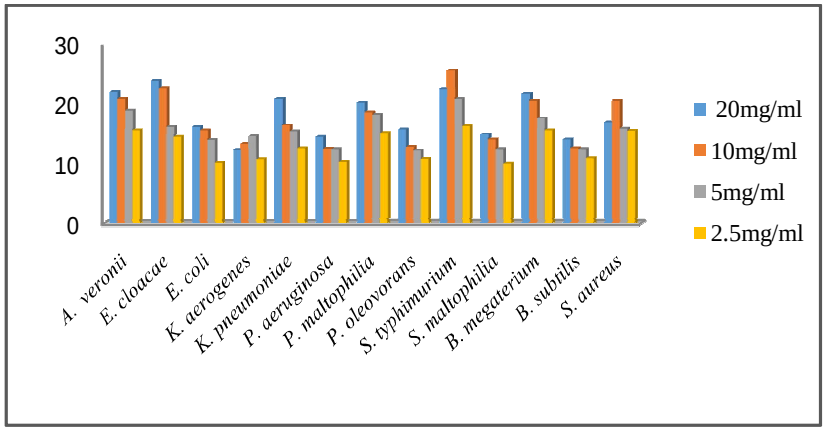


Figure 3. Antimicrobial activity of chloroform of *G. superba* tuber.

Table 4. Antimicrobial activity of hexane extract of *G. superba* tuber

Organism	NCIM Acc. No	Hexane extract mg mL^{-1}			
		20	10	5	2.5
A. veronii	5621	14.3±1.3	13.3±0.65	11.7±1.25	10.2±0.8*
E. cloacae	2164	12.7±0.8	10.8±0.7	10.5±0.4	10.1±0.2
E. coli	2068	15.2 ±0.7	12.9 ±0.3	10.7±0.6	10.0±0.2
K. aerogenes	2239	16.8±1.3	12.7±1.0	12.0 ±0.2	10.5±0.4
K. pneumoniae	2707	12.0 ±0.5	14.4 ±1.0	16.2±1.0	10.3± 0.9
P. aeruginosa	3037	12.6 ±0.7	12.0 ±0.9	10.5 ±0.8	10.2±0.6
P. maltophilia	2866	20.4±0.9	16.3±0.7	12.5±0.2	12.0±0.9
P. oleovorans	2867	20.0±1.0	18.9±0.3	16.4±0.1	14.2±0.5
S. typhimurium	2501	20.4± 0.4	22.7±0.6	20.1±0.5	18.0 ± 0.4
S. maltophilia	5625	12.8±0.3	11.3±0.8	10.8±0.4	10.2±0.7
B. megaterium	2052	14.5 ± 0.9	16.0 ±1.6	12.5±0.8	10.3±0.9
B. subtilis	2920	12.6 ± 0.4	11.3 ± 0.8	10.9 ±0.7	10.2±0.5
S. aureus	2079	30.1±0.8	25.3±0.2	22.6±1.0	12.9±0.2

* Zone of inhibition in mm

The low inhibitory zone 10.0±0.2 showed in 2.5 mg mL^{-1} concentrations against *E. coli*, 10.5±0.4 mm in 5 mg mL^{-1}, 10.8±0.7 mm in 10 mg mL^{-1} against *E. cloacae*; 12.0 ±0.5 mm in 20 mg mL^{-1} against *K. pneumoniae.* The antimicrobial activity of the hexane extracts expresses better activity against gram-positive organism than the negative organism. The hexane extract showed

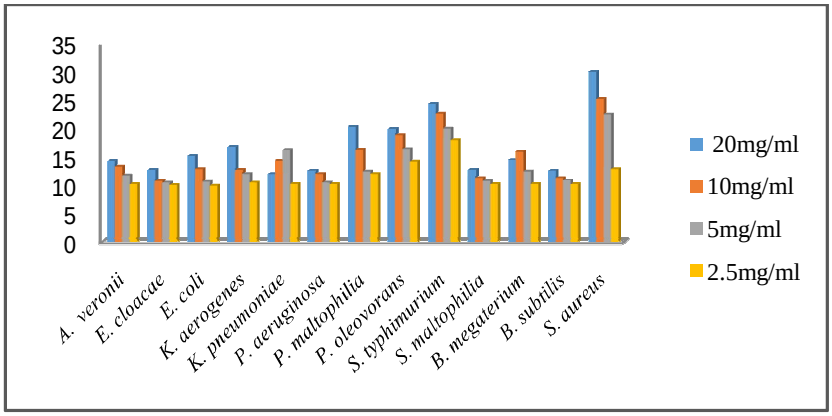


Figure 4. Antimicrobial activity of hexane extract of *G. superba* tuber.

moderate activity against all tasted organism. The pathogen *K. pneumoniae* shown more inhibitory in low dose of the extract (16.2±1.0 mm in 5 mg mL^{-1}) compare to the high dose level (12.0 ±0.5 in 20 mg mL^{-1} and 14.4 ±1.0 in 10 mg mL^{-1})

The organism *B. subtilis* activity was predominantly inhibited by aqueous extracts compare to all other extracts. It is denoted that the aqueous extract is having the active principle for controlling the *B. subtilis* infection compared to other extracts (Table 5, Figure 5).

Table 5. Antimicrobial activity of *G. superba* tuber extracts vs. standard antibiotics

Organism	NCIM Acc. No	Aqueous	Methanol	Chloroform	Hexane	Ampicillin 10 mcg	Tetracycline 30 mcg
A. veronii	5621	15.2±0.8	15.9±0.92	22.0 ±0.6	14.3±1.3	-	18±0.7*
E. cloacae	2164	-	20.6±0.4	23.9± 0.7	12.7±0.8	10±0.6	25±0.4
E. coli	2068	-	15.2±1.4	16.2±0.3	15.2 ±0.7	12±0.7	27±0.2
K. aerogenes	2239	15.0±0.7	20.0±0.7	14.6±0.4	16.8±1.3	12±0.2	24±0.8
K. pneumoniae	2707	12.8±0.8	18.3±0.8	20.9±0.6	16.2±1.0	10±0.3	23±0.5
P. aeruginosa	3037	13.6±0.6	15.5±0.8	14.5±0.9	12.6 ±0.7	-	26±0.3
P. maltophilia	2866	14.7±0.6	25.5±1.0	20.2±0.8	20.4±0.9	10±0.9	21±0.7
P. oleovorans	2867	12.4±0.7	22.6±0.4	15.7±0.3	20.0±1.0	15±0.4	20±0.9
S. typhimurium	2501	12.4±0.4	14.4±0.9	25.6±0.1	22.7±0.6	10±0.6	24±0.2
S. maltophilia	5625	14.3± 0.2	30.2±0.1	14.9±0.6	12.8±0.3	11±0.7	30±0.6
B. megaterium	2052	15.3±0.4	22.6±1.4	21.7 ±1.2	16.0 ±1.6	12±0.8	28±0.5
B. subtilis	2920	15.7±0.8	15.3±0.7	14.1±0.9	12.6 ± 0.4	12±0.5	19±0.6
S. aureus	2079	16.8±0.2	22.3± 0.5	20.6±0.7	30.1±0.8	10±0.3	24±0.9

* Zone of inhibition in mm

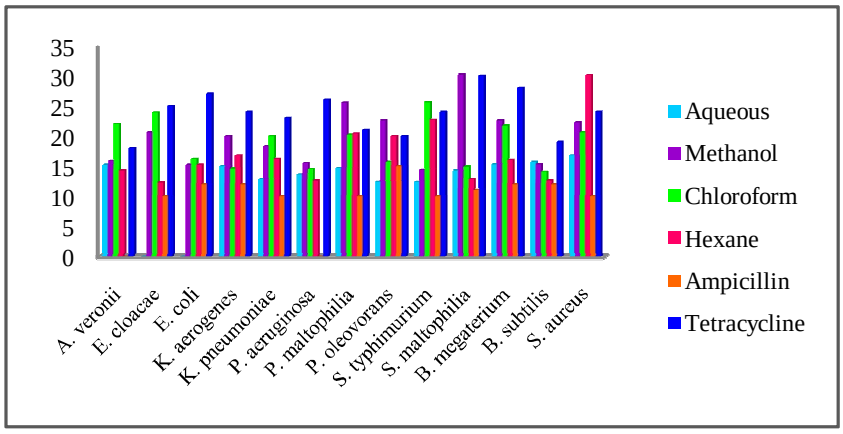


Table 5. Antimicrobial activity of *G. superba* tuber extracts vs. standard antibiotics

Similarly, Methanol extracts shown better inhibitory activity against *K. aerogenes, P. aeruginosa, P. maltophilia, P. oleovorans, S. maltophilia* among the tested organism. Accordingly, the methanol extracts have some active secondary metabolites to control these organisms than other tested organisms particularly. The chloroform extracts of the tuber are shown better antimicrobial activity against *A. veronii, E. cloacae, E. coli, K. pneumonia, S. typhimurium* compare to other tester organisms because the chloroform extracts having some active metabolite to contribute the antimicrobial activity. *S. aureus* showed maximum inhibitory effect only in hexane extract than other tested *G. superba* extracts because the extract containing phytochemicals to inhibit the activity of *S. aureus* compared to other organisms.

Medicinal plants are the primary sources of new drug and may initiate an alternative to the usual medicines. The therapeutic and aromatic plants are used on a vast, full scale in medicine against drug-resistant bacteria [**17**]. The *G. superba* is a popular ethnomedicine in Indian system of medicine for its colchicine content which is used to treat arthritis, acute gout, an antidote to snake bite, infertility, intestinal worms, laxative, skin parasites, skin problems, spines and treatment of cancer. phytochemical studies of tubers having colchicines, glycoside, gloriosine, flavonoids, tannins, alkaloids, 3-odemethylcolchicine- 3-o-α-d-glucopyranoside, 1,2- didemethyl colchicine, β and γ lumicolichicines, β sitosterol, glucoside, 2,3-didemethyl colchicine, luteolin, n-formyl deacetyl colchicines, colchicocide, tannins, superbine, 2-hydroxy-6-methoxy benzoic and salicylic acid [**9**], [**18**]. In our study the phytochemicals of tuber having less activity against *E. coli,* but this study is disagreement to earlier port said that the better inhibitory zone observed against *E. coli* because of the influence of phytochemical from the tuber [**19, 20**]. The antimicrobial activity of the extracts depends on a lot of factors like the binding capacity, chelation of iron and proteins of the bacterial cell membranes and antibacterial mechanisms of the phytochemicals [**21**]. This study is controversy to earlier report, state that the antibacterial activities of 75% methanol extract from *A. paniculata leaves* were observed only against the *S. aureus* [**22**]. In our study, the low concentration of methanol, chloroform, and hexane extracts showed a better zone of inhibition against the tested pathogen. This result is similar to the earlier of many researchers [**23-27**]. All the extracts of *G. superba* to control both gram-positive and negative organism this study positively correlated with earlier observation in *Andrographis paniculata, Begonia malabarica, Swertiacorym bosa, Drynari aquercifolia* [**28-33**].

The naturally arising alkaloids have nitrogenous compounds that institute the basic phytochemicals of flowering plants. alkaloids are formed as metabolic products and have described being accountable for pharmacological value [**34**]. Alkaloids have identified in the extracts or compounds that have been documented to possess medicinal properties and to promote health effects [**35, 36**]. Glycosides are served as defense mechanisms against predation by microbes, insects, and herbivores [**37**]. These compounds are served as essential drugs, which help the body to fight microbial infections [**38**]. Tannins have been used traditionally for protection of wound on surfaces of the mouth and treatment of catarrh, diarrhea, and hemorrhoids. Plant tannins also accepted for their pharmacological properties [**39**]. in the assessment of the previous and present results, it is clear that the plant maintains the antimicrobial property of the aqueous, hexane, chloroform and methanol extracts of the tuber of *G. superba.*

The antimicrobial effect of the raw tuber extracts is better than the standard antibiotic drugs ampicillin and tetracycline. The ampicillin showed low inhibit zone against the entire tested microorganism and did not show any activity against *A. veronii* and *P. aeruginosa.* tetracycline had shown the better inhibitory effect against *E. cloacae, E. coli, K aerogenes, K. pneumoniae, P. aeruginosa, B. megaterium, B. subtilis* than the crude extract of *G. superba.* the present study denoted that tuber has an enormous active principle to give the more antimicrobial effect than the standard abiotic disc ampicillin, tetracycline and to be isolated the secondary metabolite and also to be developed new drugs against *A. veronii, P. maltophilia, P. oleovorans, S. typhimurium, S. maltophilia,* and *S. aureus* infections.

APPLICATION

The present study denoted that tuber has an enormous active principle to give the more antimicrobial effect than the standard abiotic samples.

CONCLUSION

The aqueous and solvent extract of tuber showed antimicrobial activity against both gram-positive and gram-negative bacteria. The tuber contains alkaloid, flavonoid, terpenoids, glycosides, phenol, and tannin and these phytochemicals may be responsible for controlling the microbial infections. The present study denoted that tuber has an enormous active principle to give the more antimicrobial effect than the standard a biotic disc ampicillin, tetracycline and to be isolated the secondary metabolites and also to be developed new drugs against *A. veronii*, *P. maltophilia*, *P. oleovorans*, *S. typhimurium*, *S. maltophilia*, and *S. aureus* infections.

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