

## INVITRO ANTIMICROBIAL EFFECTS OF GOMPHOCARPUS PURPURASCENS A. RICH AGAINST STANDARD AND CLINICALLY ISOLATED MICROORGANISMS

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### Abstract

The expanding bacterial resistances to antibiotics have become a growing concern worldwide. *Gomphocarpus purpurascens* is one of the indigenous traditional medicinal plants in Ethiopia. The objective of this study was to evaluate antimicrobial effect of *G. purpurascens* against standard and clinically isolated microorganisms. The *G. purpurascens* plant leaves and root was collected from Gondar Zuria Woreda was shade dried and powdered using wooden-made mortar and pestle. The powdered leaves were extracted by using ethanol, methanol and acetone. The extracts from the leaves and root barks with different concentrations were tested for their antimicrobial activities against selected microorganisms. Minimum Inhibitory Concentration (MIC) and Minimum Bacterial Concentration (MBC) were determined against test organisms. The most sensitive bacterial species to the highest concentration of the extract from the leaf was clinical *S. aureus* strain with inhibition zone diameter of 17.66 mm; whereas the least sensitive bacterial species was clinical isolate of *E. coli* with inhibition zone diameter of 6.54±0.13 mm. These research findings suggest that *G. purpurascens* plants may contain antibacterial and antifungal compounds. These plants can be a potential source for the development of antibacterial and antifungal drugs.

**Key words/Phrases:** Antimicrobial, disk diffusion, *Gomphocarpus purpurascens*, MIC, MBC, inhibition zone.

## Introduction

The expanding bacterial resistance to antibiotics has become a growing concern worldwide (Gardam, 2000). The problem of microbial resistance has become a global issue of concern, as about 70% of the bacteria that cause infections in hospitals are resistant to at least one of the antibiotic most commonly used for treatment (Bisht *et al.*, 2009). In Ethiopia botanicals are traditionally used to cure some of the diseases in rural and some urban areas. However, the effects from most botanicals are not determined experimentally.

The increasing failure of chemotherapeutics and antibiotic resistance exhibited by pathogenic microbial infectious agents necessitates the screening of medicinal plants for their potential antimicrobial activity (Njimoh *et al.*, 2015). Increasing bacterial resistance is prompting a resurgence in research of the antimicrobial role of herbs against resistant strains (Alviano and Alviano, 2009; Hemaiswarya *et al.*, 2008). A vast number of medicinal plants have been recognized as valuable resources of natural antimicrobial compounds (Mahady, 2005). Medicinal plant extracts offer considerable potential for the development of new agents effective against infections currently difficult to treat (Iwu *et al.*, 1999).

Crude extracts of some medicinal plants in Ethiopia such as *Jasminum abyssinicum*, *Solanecio gigas* and *Lagenaria siceraria* showed strong antimicrobial activities that can serve as sources of effective drugs development against diseases causing microorganisms (Abera, 2014). Ethiopia is one of the countries has diversified flora which can contribute in producing effective antimicrobial drugs. Jansen (1981) worked out detailed research on 25 species, condiments and medicinal plants and listed 406 plant species which have either one or more than one of the above applications in Ethiopia. However, there is no much studies conducted on the *G. purpurascens*. Therefore this study was initiated to evaluate the *invitro* antibacterial activity of crude leaf and root bark extractions of *G. purpurascens* against clinically important selected microorganisms.

## Materials and Methods

### *Study area*

The study was conducted in the Department of Biology at University of Gondar, Tewodros Campus. The plant materials were collected from “Maraki” Campus which is protected and natural environment with an elevation of 2174 meters above sea level and also Azezo locality near to the northern part of Gondar Air Port. The annual average rainfall, relative humidity and monthly average temperature of the areas where the plants collected for the last 22 years average was 1216mm, 49.28% and 20.42<sup>0</sup>C, respectively (Ethiopian National Meteorology Agency, 2010).

### *Plant materials*

The plant used for the study was *G. purpurascens* which was locally called as “Tefreina”. The identity of the plant was confirmed at Ethiopian Flora Project Herbarium, Addis Ababa University. From each locality selected for plant collection, five healthy, disease free and young plant parts were collected randomly. The collected plant parts were kept in plastic bags and brought to the laboratory for subsequent processing.

### *The study design*

The study was cross sectional laboratory based experimental design. The different plant parts such as leaves, stem and root bark was washed thoroughly 2-3 times with running tap water and once with sterile distilled water. Then the plant parts were air-dried on sterile blotter under shade and powdered using wooden-made mortar and pestle. According to Dhanani *et al.* (2017), the powdered plant materials were mixed with 97% ethanol, 99.5% methanol and 99.5% acetone individually and kept in shaker for 72 hours and filtered using Whatman No 1 filter paper. After shaking, the filtrate was transferred into flask and solvents removed by using rotary evaporator. Water from the extract was further evaporated on water bath and the solvent free residue was used for further investigation. The extracts were stored in a refrigerator at 4<sup>0</sup>C until use. Consequently, extracts for antimicrobial activity were monitored against the selected standard and clinically important human pathogens using disk diffusion method and the diameter of zone of inhibition was measured. Minimum Inhibitory Concentration (MIC) was determined by two fold broth dilution method. Finally, Minimum Bactericidal Concentration (MBC) values were determined by sub- culture from the MIC value according to Ramli *et al.* (2016). Antimicrobial

activities of each extract were evaluated from their mean diameters of inhibition zones from three replications.

### ***Preparation of Inoculum***

Clinical isolates of bacterial pathogens such as *E. coli*, *P. aeruginosa*, and *S. aureus* as well as standard strains of *E. coli* (ATCC25922), *S. aureus* (ATCC25923) and *S. pneumonia* (ATCC63) as well as *C. albicans* were collected from Biomedical and Laboratory Sciences Research Center, University of Gondar. These microorganisms were taken to Microbiology Laboratory by using nutrient agar slant and preserved at 4<sup>0</sup> C until used for subsequent experiments. The turbidity of actively growing bacterial suspension was adjusted to match the turbidity standard of 0.5 McFarland units using following the method of Willey *et al.* (2008). These actively growing bacteria were inoculated on Muller Hinton Agar medium.

### ***Antibacterial and antifungal Bioassay***

Standard disc diffusion method described by Kirby-Bauer (Willey *et al.*, 2008) was used for this study. Test discs (6 mm diameter) was prepared and dipped in plant extracts. The crude extracts from each part was tested at 150, 300 and 600 mg/ml. Tetracycline and Ketoconazole was used as standard antibacterial and antifungal drugs (positive control) at 0.025 mg/disc and 25 mg/disc, respectively. For negative control discs was impregnated with 80% methanol. The discs were placed on the cultured bacteria and fungus using sterile forceps by leaving equal distances between two nearby discs. Finally, the cultured bacteria and fungus was incubated for 24 hours at 37°C and 24 -72 hours at 27°C, respectively. The diameters from each zone of inhibition as indicated by clear area were measured as devoid of growth of bacteria and fungi.

### ***Determination of Minimum Inhibitory Concentration***

Minimum Inhibitory Concentrations (MIC) of crude extracts was performed using two fold broth dilution methods (Abew *et al.*, 2014). The extract solution (600 mg/ml) was serially diluted with nutrient broth as 1:2, 1:4, 1:8, and 1:16 to bring 300 mg/ml, 150 mg/ml, 75 mg/ml and 37.25 mg/ml concentrations, respectively and 20µl of a standard suspension of the test organism were added to each concentration of the extract. Two test tubes containing nutrient broth without antimicrobial agent was added in each test. One of these tubes was inoculated with the test organism; the other was not being inoculated and served as a check for media sterility. The broth

tubes were incubated at 37°C for 24 hr. The lowest concentration, at which there is no turbidity, was considered as MIC value of the extract.

### ***Determination of Minimum Bactericidal Concentration***

A Minimum Bactericidal Concentration (MBC) is the lowest concentration of an antibiotic needed to kill a microorganism (BSAC, 2010). The MBC was determined by sub-culturing 20 µl of the test dilutions from MIC tubes on to fresh nutrient agar plates incubating for 24 hr. The lowest concentration that kills the entire bacterial colony on the plates was recorded as MBC.

### ***Data Analysis***

The data collected from all the experiments were entered in to Microsoft excel spread sheet and analyzed using Microsoft Excel Spread Sheet ver. 2010 and PAST ver.1.34 (Hammer *et al.*, 2005). The diameter zone of inhibition was expressed as mean  $\pm$  standard deviation (M  $\pm$  SD). One-way analysis of variance (ANOVA) was used for determination of the antimicrobial susceptibility test of botanicals extracted by different solvents in the specific concentration. Results were considered statistically significant at P-value  $< 0.05$ .

### **Results**

The means with standard deviations of inhibition zones diameters scored from *G. purpurascens* leaf extract on bacterial and fungal species with three replication was presented in Table1. The test parts of *G. purpurascens* showed positive activities as evidenced by a clear zone of inhibition against the tested bacteria and fungus ( $P < 0.05$ ). Though the response was not uniform, leaf extracts of the plant showed activity against the tested bacterial strains and fungus used in this assay. Results obtained in the present study revealed that the tested plant part extracts possess potential antibacterial and antifungal activity against all tested organisms. The highest antibacterial activity was 14.51mm (in acetone extract) in standard *S. aureus* and least activity was recorded in standard *E. coli* measured 6.09 mm (in ethanol extract) in the lowest concentration (150 mg/mL). The sensitive bacterium to the highest concentration (600 mg/mL) of the leaf extract was shown on clinical *S. aureus* with inhibition diameter of 17.66 mm (in methanol extract), followed by standard strain *S. aureus* with inhibition diameter of 17.45 mm (in acetone extract). On the contrary, clinical *E. coli* (in acetone extract) and clinical *S. aureus* (in ethanol extract) were the least sensitive to the leaf extract with inhibition diameters of 6.54 mm

and 6.60 mm, respectively. *C. albicans* was found susceptible to the lowest concentration (150 mg/mL) with inhibition diameter of 8.01 mm (in methanol extract) but least was found from ethanol extract with inhibition diameter of 6.34 mm.

Results obtained in the present study from root bark extracts revealed that except for *C. albicans* remaining microorganism observed potential antibacterial activity. The mean zone of inhibition at different concentration levels of root bark extracts of *G. purpurascens* increased proportionally as concentration increased which was statistically significant ( $p < 0.005$ ) as shown from Table 2. The minimum concentration (150 mg/mL) of ethanol root bark extracts of *G. purpurascens* exhibit highest antibacterial activity against clinical isolate of *P. aeruginosa* (11.34 mm) and lowest in standard strain of *E. coli* (8.6 mm). Root bark and leaf extract of *G. purpurascens* showed almost similar antibacterial activity against all the tested bacteria. Minimum concentration (150 mg/mL) of leaf extract of *G. purpurascens* showed highest activity against standard strains of *S. pneumonia* and *S. aureus* with inhibition diameter of 9.67 mm and 9.51 mm, respectively while lowest activity were observed in standard strain of *E. coli* (6.09 mm) and clinical isolate of *S. aureus* (6.15 mm). Root bark extract of this plant shown maximum activity against clinical *P. aeruginosa* (11.34 mm) followed by standard strains of *S. aureus* and *S. pneumonia* with inhibition diameter of around 10.32 mm and the minimum activity were observed in standard strain of *E. coli* with inhibition zone diameter of 8.6 mm. Root bark extract showed greater activity when compared with the leaf extract of test plant extracts.

Table 1. Diameter of inhibition zone at different concentrations of leaf extracts of *Gomphocarpus purpurascens* obtained using three different solvents (disc diffusion method)

Solvent	Extract	Extract Conc.(mg/mL)	Diameter of inhibition zone (mm)						Fungal species
			Bacterial species						
			<i>E. coli</i> *	<i>E. coli</i> **	<i>P. aeruginosa</i> *	<i>S. aureus</i> *	<i>S. aureus</i> **	<i>S. pneumonia</i> **	<i>C. albicans</i>
Acetone		150	6.35±0.13	6.33±0.35	7.01±0.01	9.91±0.15	14.51±0.15	11.94±0.40	6.67±0.15
		300	6.34±0.24	6.4±0.40	7.07±0.07	12.53±0.02	15.01±0.15	12.04±0.036	6.71±0.015
		600	6.54±0.13	6.96±0.72	7.36±0.30	13.92±0.02	17.45±0.20	13.70±0.020	7.47±0.025
Ethanol		150	6.77±0.45	6.09±0.18	9.01±0.01	6.15±0.20	9.51±0.10	9.67±0.015	6.34±0.04
		300	7.17±0.15	6.27±0.25	9.1±0.01	6.30±0.15	11.13±0.25	10.93±0.03	6.63±0.025
		600	7.41±0.10	6.90±0.40	9.77±0.15	6.60±0.20	12.70±0.15	13.15±0.35	6.64±0.04
Methanol		150	6.68±0.98	9.19±0.07	9.80±0.01	13.93±0.025	7.1±0.01	10.74±0.036	8.01±0.015
		300	6.89±0.90	10.27±0.15	12.27±0.25	14.01±0.015	7.90±0.15	14.12±0.01	12.26±0.02
		600	7.73±0.35	13.20±0.10	13.80±0.10	17.66±1.17	8.21±0.01	14.80±0.025	13.79±0.01
Tetracycline		0.025	10.78±1.29	13.09±1.45	17.71±2.26	20.26±1.27	25.65±2.49	26.68±2.4	
Ketoconazole		25							7.87±2.18
Negative control			-	-	-	-	-	-	-

Conc. Concentration, \*Clinical isolates, \*\* Standard strains, Standard drugs-Tetracycline (0.025 mg/ml) for Bacterial strains and Ketoconazole (25 mg/ml) for *Candida albicans*, Values are mean ± standard deviation of triplication (Mean ± S.D)

Table 2. Antimicrobial activity of leaf and root bark extracts of *G. purpurascens* obtained using ethanol as a solvent at three different concentrations using disk diffusion methods (inhibition zone in mm) comparing with positive controls

Test organism	Leaf extract in mg/mL				Methanol	Positive controls	Root bark extract in mg/mL				Methanol	Positive controls
	150	300	600	$\bar{X} \pm SD$			150	300	600	$\bar{X} \pm SD$		
<i>E. coli</i> *	6.77±0.45	7.17±0.15	7.41±0.10	7.12±0.23	-	10.78±1.29	10.53±0.03	12.17±0.12	17.52±0.02	13.41±0.06	-	9.34±0.40
<i>E. coli</i> **	6.09±0.18	6.27±0.25	6.90±0.40	6.42±0.28	-	13.09±1.45	8.60±0.10	8.94±0.04	9.42±0.02	8.99±0.05	-	11.98±0.02
<i>P. aeruginosa</i> *	9.01±0.01	9.1±0.01	9.77±0.15	9.30±0.06	-	17.71±2.26	11.34±0.04	12.27±0.15	18.03±0.03	13.88±0.07	--	15.43±0.21
<i>S. aureus</i> *	6.15±0.20	6.30±0.15	6.60±0.20	6.35±0.18	-	20.26±1.27	9.01±0.02	10.80±0.01	10.86±0.01	10.22±0.01	-	19.70±0.10
<i>S. aureus</i> **	9.51±0.10	11.13±0.25	12.70±0.15	11.11±0.17	-	25.65±2.49	10.32±0.02	11.33±0.15	14.71±0.01	12.12±0.06	-	14.30±0.20
<i>S. pneumonia</i> **	9.67±0.02	10.93±0.03	13.15±0.35	11.25±0.13	-	26.68±2.4	10.16±0.01	11.27±0.02	14.96±0.01	12.13±0.01	-	26.40±0.36
<i>C. albicans</i>	6.34±0.04	6.63±0.03	6.64±0.04	6.54±0.04	-	7.87±2.18	-	-	-	-	-	7.87±2.18

$\bar{X}$  Grand mean,\*Clinical isolates; \*\* Standard strains; Values are mean ± standard deviation of triplication (Mean± S.D), Positive controls-Tetracycline (0.025 mg/ml) for Bacterial strains and Ketoconazole (25 mg/ml) for *C. albicans*; and No inhibition zone



The Minimum Inhibitory Concentration (MIC) of the leaf extracts of *G. purpurascens* against the tested bacteria ranged from 37.5 mg/mL to 300 mg/mL for each solvent (Table 3). The results revealed variability in the inhibitory concentration of each extract for given microorganisms. The MIC values of acetone and methanol extracts ranged from 150 mg/mL to 75 mg/mL with the highest MIC values compared to ethanol extracts fraction. Ethanol extracts had MIC values ranged from 150 mg/ml (on standard *E. coli* (ATCC25922) and standard *S. pneumonia* (ATCC63) to 37.25 mg/ml (on standard *S. aureus* (ATCC 25923) with the lowest MIC values compared to the other extracts fraction. Among the bacterial isolates and standard *S. aureus* (ATCC 25923) were highly susceptible to the smallest MIC value of ethanol extracts followed by clinical strain *P. aeruginosa*, clinical *S. aureus*, clinical *S. pneumonia* which were inhibited by the concentration of 75 mg/ml on all extracts . Isolates of standard *E. coli* (ATCC25922) were the least inhibited tested organisms having MIC values of 150 mg/ml at all extracts. *C. albicans* was highly susceptible to the methanolic extract with MIC value of 75 mg/ml but less inhibited in the ethanolic extract of the plant with MIC value of 300 mg/ml.

Table 3. The MIC values of *G. purpurascens* leaf extracts against test organisms using two fold broth dilution methods.

Test organism	Plant leaf extract (mg/ml)		
	Acetone extract	Ethanol extract	Methanol extract
<i>E. coli</i> *	150	75	150
<i>E. coli</i> **	150	150	150
<i>P. aeruginosa</i> *	75	75	150
<i>S. aureus</i> *	150	75	75
<i>S. aureus</i> **	75	37.5	150
<i>S. pneumonia</i> **	75	150	75
<i>C. albicans</i> **	150	300	75

\*Clinical isolates, \*\* Standard strains

#### *Minimum bactericidal concentrations of leaf extract*

Minimum bactericidal concentrations (MBCs) of each extract was expressed as the lowest dilution level of the extract needed to completely inhibit bacterial growth. Acetone extract of leaf fractions showed MBC values which ranged from 150 mg/mL (against clinical isolate and

standard strains of *E. coli* (ATCC25922), clinical *P. aeruginosa* and standard *S. aureus* (ATCC 25923) to 75 mg/mL (against clinical isolate of *S. aureus* and standard *S. pneumonia* (ATCC63) (Table 4). Similarly methanol extract fractions showed MBC values ranged from 150 mg/mL (against clinical isolates of *E. coli*, *S. aureus*, standard strains of *E. coli*, (ATCC25922) *S. aureus* and *S. pneumonia* (ATCC63) to 75 mg/mL (against clinical isolate of *P. aeruginosa*). However, the MBC values of ethanol extract fractions ranged from 300 mg/mL (against standard strain of *E. coli* (ATCC25922) to 75mg/mL (against standard strain of *S. aureus* (ATCC 25923), the remaining tested organisms MBC value is 150 mg/mL). Within the bacterial isolates, standard *E. coli* (ATCC25922) was found less susceptible to the extract prepared using ethanol having MBC value of 300 mg/ml.

Table 4. The minimum inhibitory concentration (MBC) of *G. purpurascens* leaf extracts against test organisms

Test organism	Plant extract (mg/ml)		
	Acetone extract	Ethanol extract	Methanol extract
<i>E. coli</i> *	150	150	150
<i>E. coli</i> **	150	300	150
<i>P. aeruginosa</i> *	150	150	75
<i>S. aureus</i> *	75	150	150
<i>S aureus</i> **	150	75	150
<i>S pneumonia</i> **	75	150	150

\*Clinical isolates, \*\* Standard strains

#### *Minimum inhibitory concentration and Minimum Bacterial Concentration of root bark extract*

The MIC value of root bark extracts of *G. purpurascens* against the tested bacteria was ranged from 37.5 mg/mL to 300 mg/mL for the solvent ethanol (Table 5). The results showed variability in the inhibitory concentration of each extract for given microorganisms. Ethanol extracts had MIC values ranged from 75 mg/mL (on clinical isolates of *E. coli* and *S. aureus* as well as against standard *S. pneumonia* (ATCC63) to 37.5 mg/ml (on standard *S. aureus* (ATCC 25923) and clinical isolate of *P. aeruginosa*) with the lowest MIC values compared to the other extracts fractions. Within the bacterial isolates standard *S. aureus* (ATCC 25923) and clinical *P. aeruginosa* were highly susceptible to the smallest MIC value of ethanol extracts which was 37.5

mg/mL while the least susceptible were clinical strain of *E. coli*, *S. aureus*, and standard *S. pneumonia* (ATCC63) which was inhibited by the concentration of 75 mg/mL.

Table 5. Minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of *G. purpurascens* root bark extracts against test organisms.

Test organism	MIC (mg/mL)	MBC (mg/mL)
<i>E. coli</i> *	75	37.5
<i>E. coli</i> **	-	-
<i>P. aeruginosa</i> *	37.5	37.5
<i>S. aureus</i> *	75	75
<i>S. aureus</i> **	37.5	75
<i>S pneumonia</i> **	75	75
<i>C. albicans</i> **	-	-

\*Clinical isolates, \*\* Standard strains, - No MIC Value

Minimum bactericidal concentrations (MBCs) of ethanol extract were expressed as the lowest dilution level of the extract needed to completely inhibit bacterial growth. Ethanol extract root bark fractions showed MBC values which ranged from 75 mg/mL (against clinical isolates and standard strains of *S. aureus* (ATCC 25923), and standard *S. pneumonia* (ATCC63)) to 37.5 mg/mL (against clinical isolate of *E. coli* and clinical isolate *P. aeruginosa*) (Table 5). Among the examined bacterial isolates, clinical isolates of *E. coli* and *P. aeruginosa* were found more susceptible to the root bark extract of *G. purpurascens* prepared using ethanol with MBC value of 37.5 mg/mL. Clinical isolate of *S. aureus* and standard strains of *S. aureus* and *S. pneumonia* was the least susceptible with MBC value of 75 mg/mL.

## Discussion

The type and level of biological activity exhibited by any plant material depends on many factors, including the plant part, geographical source, soil conditions, harvest time, moisture content, drying method, storage conditions, and post-harvest processing. For example, the relatively high temperatures that can be generated during tissue grinding can denature chemical constituents and the extraction solvent, time period, and temperature can affect the level and composition of secondary metabolites extracted from plant tissues (Wendakoon *et al.*, 2012).

The plant chosen for this study was commonly used for treating infectious diseases such as itching skin in herbal therapy (Anteneh and Nigussie, 2014).

The result of this study showed that acetone, ethanol and methanol extracts of leaf and ethanolic extract of root bark of *G. purpurascens* showed varied antibacterial and antifungal activity against the tested organisms, except for root bark extract of ethanol that showed no activity against *C. albicans*. This result suggests that the extracts of *G. purpurascens* tested parts are broad spectrum in their activities since it showed activity against Gram positive and Gram negative bacteria. This correlates with the observation of previous researches they have reported that plants have antimicrobial activity (Iwu *et al.*, 1999; Cowan, 1999; Rios and Recio, 2005; Cos *et al.*, 2006).

In the present study, the lowest concentration (150 mg/mL) of *G. purpurascens* leaves (acetone extract) and root bark (ethanol extract) showed strongest antibacterial activity against standard strain of *S. aureus* and clinical isolate of *P. aeruginosa* with mean zone of inhibition diameter of  $14.51 \pm 0.15$  mm and  $11.34 \pm 0.04$  mm, respectively. The extracts from *G. purpurascens* leaf and root bark had by far more antimicrobial effect as compared with *Solanum ferox* plant extract applied on pathogenic bacteria of *Aeromonas hydrophila* and *Pseudomonas* spp. (Hardi *et al.*, 2016). But it was not effective as some plants' extracts were effective from Vu *et al.* (2016) report. Comparing with leaf extract from *Syzygium polynathum* (Ramli, *et al.*, 2016) leaf extract from the current study showed similar result whereas its root extract showed the formation of larger inhibition zone than both *S. polynathum* leaf extract and the standard antibiotics used in their study for most bacterial strains. Sakha *et al.* (2019) reported similar inhibition zone result on the same bacterial strain using different plant species leaves extracts. The result of the present study is in line with the findings of Belayneh and Bussa (2014) on screening of the antimicrobial activities of some plants used traditionally in Ethiopia for the treatment of skin disorders and Ethno medicinal plants used to treat human ailments in the prehistoric place of Harla and Dengego valleys, respectively although their result was not supported by *invitro* experiments. As result, this study provides base line data on *invitro* antimicrobial activities of *G. purpurascens*.

In the present study, Gram-positive bacteria were more sensitive than the Gram-negative bacteria. This was also reported by several researchers (Vu, *et al.*, 2016; Ramli *et al.*, 2018). The reason for higher sensitivity of the Gram-positive bacteria compared to Gram-negative bacteria

could be ascribed to their differences in cell wall constituents and their arrangement (Ramli *et al.*, 2016). The Gram-positive bacteria contain a peptidoglycan layer, which was an ineffective permeability barrier while Gram-negative bacteria was surrounded by an additional outer membrane carrying the structural lipopolysaccharide components, which makes it impermeable to lipophilic solutes and porins constitute a selective barrier to the hydrophilic solutes (Nikaido, 2003).

In the present investigation, most of the MBC and MIC values of the plant extracts were almost similar and this statement is agreed with Taye *et al.* (2011). Among the bacterial isolates standard *S. aureus* (ATCC 25923) was highly susceptible to the smallest MIC value of ethanol extracts followed by clinical strain *P. aeruginosa*, clinical *S. aureus*, clinical *S. pneumonia* which was inhibited by the concentration of 75 mg/ml on all extracts. Isolates of standard *E. coli* (ATCC25922) was the least inhibited tested organisms having MIC values of 150 mg/ml at all extracts.

## Conclusion

From the results obtained, it is evident that *G. purpurascens* leaves and root possesses potential inhibitory activity against human pathogens (bacteria and fungi). Standard *S. aureus* (ATCC 25923) was highly susceptible to the least MIC value of leaf ethanol extracts followed by clinical strain *P. aeruginosa*, clinical *S. aureus*, clinical *S. pneumonia* which were inhibited by the concentration of 75 mg/ml on all leaf extracts. Isolates of standard *E. coli* (ATCC25922) was the least inhibited tested organisms having MIC values of 150 mg/ml at all extracts. Within the bacterial isolates, standard *E. coli* (ATCC25922) was found less susceptible to the extract prepared using ethanol having MBC value of 300 mg/ml. Clinical isolates of *E. coli* and *P. aeruginosa* were found more susceptible to the root bark extract of *G. purpurascens* prepared using ethanol with MBC value of 37.5 mg/mL. Generally, *G. purpurascens* root bark extract showed maximum antibacterial inhibition effect than leaf extract. This *invitro* study results demonstrates that the plant parts may be contain potential antibacterial compounds. Further isolation of the compounds with appropriate clinical trials will be important to develop antimicrobial drugs for the treatment of infectious diseases.

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