# The *in vitro* evaluation of some South African plant extracts for minimum inhibition concentration and minimum bactericidal concentration against selected bacterial strains

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**ABSTRACT**
Bacterial infections are one of the world's most pressing public health problems. The major challenge in anti-bacterial treatment is the development of antibiotic resistant bacterial strains. This then reduces the number of drugs available for treating bacterial infections. In this study, antimicrobial activity of nine South African crude plant extracts were investigated for the first time against *Escherichia coli* (*E.coli*), *Pseudomonas aeruginosa* (*Ps aeruginosa*), *Staphylococcus aureus* (*S. aureus*) and *Enterococcus faecalis* (*E. faecalis*). The minimum inhibitory concentrations of the crude plant extracts were determined. Four plant extracts showed good inhibition effects against all four bacteria used which are *Lippia javanica* (0.25±0.00 to 1.13±0.29 mg/ml); *Ziziphus mucronata* leaf (0.44±0.00 to 1.00±0.00 mg/ml); *Erythrina lysistemon* (0.44±0.00 to 1.08±0.00 mg/ml) and *Schkuhria pinnata* (0.5±0.00 to 1.34±0.00 mg/ml). Similarly, gentamycin (positive control) had inhibitory effects ranged from 0.92±0.29 to 2.00±0.00 mg/ml against all four bacteria used. For the bactericidal effects, four plant extracts that showed bactericidal effects against three bacteria used *(E.coli*, *S. aureus* and *E. faecalis*). The plant extracts are *Lippa javanica* (0.25±0.00 to 1.00±0.00 mg/ml); *Ziziphus mucronata* leaf (0.25±0.00 to 2.00±0.00 mg/ml); *Erythrina lysistemon* (0.5±0.00 to 2.00±0.00 mg/ml) and *Schkuhria pinnata* (0.5±0.00 to 2.00±0.00 mg/ml. Furthermore, none of the plant extract showed bactericidal against Ps. *aeruginosa*. The results of this study suggests that four plants that demonstrated inhibition effect and bactericidal effects which supports their used in the traditional medicine treatment of various aliments.

**Keywords**: bacterial infections, antimicrobial activities, pathogens, inhibition, bactericidal.

## 1.0 INTRODUCTION

Bacterial infections are caused by pathogenic microorganisms that have the ability to cause diseases in humans (Alberts *et al*., 2002; Ryan *et al*., 2014). Not all bacteria are pathogenic, the ones that are pathogenic are responsible for causing diseases in humans. The most prominent pathogenic microorganism is *Mycobacterium tuberculosis* which is responsible for the highest tuberculosis burden in humans; and accounts for about 2 million deaths per year, mostly in sub-Saharan Africa (Chan *et al*., 2013; Ryan *et al*., 2014). Another globally important genera of pathogens are *Streptococcus* and *Pseudomonas* which are responsible for diseases such as pneumonia. Genera such as *Shigella*, *Campylobacter* and *Salmonella* are mainly responsible for foodborne illnesses. In addition to this, they can further cause infections such as tetanus, typhoid fever, diphtheria, syphilis and leprosy (Ben-Noun, 2009; Chan *et al*., 2013).

The increase of antimicrobial resistance is a global concern. It is characterised by multidrug-resistance which is mainly caused by gram-negative bacteria (Exner *et al*., 2017). This is due to genetic mutations and can also be triggered by misuse of antimicrobial drug prescriptions (Mwambete, 2009). Gram-negative strains that contribute towards multidrug-resistance have been reported and these include, *Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa, Enterobacter* spp and *Escherichia coli (*Exner *et al*., 2017). Although more attention has been focused on multi-drug resistance (MDR) associated with gram-negative bacteria, and gram-positive antimicrobial resistance are also a serious concern (Doemberg *et al*., 2017). For example, methicillin-resistant *Staphylococcus aureus* (MRSA) is a typical example that has raised increased global concern as a cause of community-acquired and healthcare-associated infection. Gram-positive organisms including bacteria of the genera *Staphylococcus*, *Streptococcus* and *Enterococcus*, are among the most common bacteria causes of clinical infection (Johnson *et al*., 2005; Klevens *et al*., 2007). The substantial increase in the drug resistance of antibiotics in recent years poses an ever-

increasing therapeutic challenges.

The organisms play a pivotal role in developing resistance to antibiotics. However, patients are also partly responsible for antibiotic resistance due to overuse, misuse, and/or lack of adherence to the prescription, failure to complete the course which has led to ever-increasing levels of resistance to antibiotic treatment (Franco *et al*., 2009; Prestinaci *et al*., 2016). Drug resistance has become particularly problematic in recent times because of the slow pace at which novel antibiotics are being discovered while antibiotic use continues to rise (Li and Webster, 2018). Due to the prevalence of microbial resistant strains, there is a need to focus on the plant species that show antimicrobial activities. The identification of plant species with antimicrobial properties is of great importance for therapeutic treatment of patients (Artizzu *et al*., 1995; Izoo *et al*., 1995; Eloff, 1998; Selvamohan *et al*., 2012).

It is important to evaluate the ability of plant extracts to inhibit the growth of known pathogenic microorganisms. The general method used by most investigators is agar diffusion assay for the determination of microbial activities (Nascimento *et al*., 2000; Al-Hussainin and Mahasneh, 2009). For the interest of this study, a sensitive and quick microplate method previously described by Eloff (1998) was used. This method is useful when investigating extracts with unknown components. Furthermore, this technique addresses shortfalls that have been observed on the agar diffusion; such as microbial effects may be inhibited by extrinsic factors or contamination (Marsh and Goode, 1994). Moreover, this method clearly distinguishes between bactericidal and bacteriostatic effects and it can determine minimum inhibitory concentration (Eloff, 1998). In addition, the minimum bactericidal concentration can be defined as the lowest concentration that can kill any visible bacterial growth (Sen and Batra, 2012).

## 2.0 METHODS AND MATERIALS

### 2.1 Plant collection and extraction

Plant species (n=9) were collected from Walter Sisulu National Botanical Gardens in South Africa, in February 2017 (**see Table 1 here below**). The voucher specimen are held at Walter Sisulu National Botanical Gardens herbarium. The plant material was air-dried in a well ventilated room. After drying, the plants were ground into fine power and stored away from light at room temperature.

**Table 1**: Nine plant species with accession numbers and voucher specimen numbers use in this study.

| NUMBER | NAME | FAMILY | PART | ACCESSION NUMBER | VOUCHER OF SPECIMEN COLLECTED | |
|---|---|---|---|---|---|---|
| | | | | | DATE | NUMBER |
| 1 | *Euclea crispa* | Ebenaceae | Leaf | 24/1982 | 11/10/1982 | 24, Behr, C.M |
| 2 | *Euclea natalensis* | Ebenaceae | Leaf | 178/1987 | 10/6/1987 | 479; Steel, B.S |
| 3 | *Schuhria pinnata* | Asteraceae | Weed | N/A | N/A | N/A |
| 4 | *Ziziphus mucronata* | Rhamnaceae | Leaf | 36/1982 | 15/10/1982 | 39; Behr, C.M |
| 5 | *Ziziphus mucronata* | Rhamnaceae | Fruit | 36/1982 | 15/10/1982 | 39; Behr, C.M |
| 6 | *Lippia javanica* | Verbenaceae | Leaf | 16/2014 | 22/1/2014 | 28; Kondlo, M |
| 7 | *Vernonia oligocephala* | Asteraceae | Leaf | 268/2013 | 12/05/2013 | 29; Hankey, A.J |
| 8 | *Clerodendrum myricoides* | Lamiaceae | Leaf | 11/1987 | 2/2/1987 | 367, Steel, B.S |
| 9 | *Erythrina lysistemon* | Fabaceae | Leaf | 21/1982 | 7/10/1982 | 22; Behr, C.M |

### 2.2 Preparation of crude extracts for biological assays

The ground plant extracts (leaves and fruit) were extracted with 90% methanol (1 g/10 ml) in clean honey jars and vigorously shaken for 3 hours. The crude extracts were filtered through Whatman No.1 filter paper and dried at room temperature under a stream of cold air. The crude extracts were reconstituted in methanol, dichloromethane and acetone at a concentration of 10 mg/ml for all assays

### 2.3 Bacterial strains

The antimicrobial activities of the plant extracts were tested against gram-negative bacteria [*Escherichia coli* (NCTC10538) and *Pseudomonas aeruginosa* (ATCC27853)] and gram-positive bacteria [*Staphylococcus aureus* (NCTC10538) and *Enterococcus faecalis* (ATCC29212)]. These bacterial strains were donated by the Pearson Institute of Higher Education, South Africa and the University of Pretoria Paraclinical Department. The bacterial strains were continuously maintained in nutrient agar and Mueller-Hinton agar sub-cultured weekly in order to ensure that fresh cultures were used each time for an inoculum preparation.

### 2.4 Determination of minimal inhibitory concentration (MIC)

Minimal inhibitory concentrations (MIC's) were determined using the microplate dilution method developed by Eloff (1998). Methanol (MeOH) crude plant extracts were re-dissolved in 50% distilled water and 50% solvent, namely MeOH, dichloromethane (DCM) and acetone (AC) to a concentration of 8 mg/ml. One hundred microliters (100 µl) of dissolved plant extract and a positive control, gentamicin (Sigma-Aldrich, Cat No, 1405-41-0) were serially diluted with distilled water in a 96 well plate. Bacterial suspension (100 µl), standardised to McFarland standard No. 0.5, was added to each well. The following concentrations of plant extracts were used in these experiments: 2, 1, 0.5, 0.25, 0.125, 0.062, 0.031 and 0.015 mg/ml respectively. Sealed plates were incubated at 37℃ for 24 hours.
To indicate growth, 40 µl of *p*-iodonitrotetrazolium violet (INT) (Sigma-Aldrich, Cat NO. 18377), was dissolved in distilled water and added to the microplate wells were incubated at 37℃ for 30-45 minutes. The presence of bacterial growth was observed as a pink/red colour, and clear wells indicated the inhibition of bacterial growth by the plant extract. The extract dissolving solvents (MeOH, DCM and AC) were used as negative controls and gentamicin served as a positive control (50 µg/ml). The assay was performed in quadruplicates and repeated three times.

### 2.5 Determination of minimal bactericidal concentration (MBC)

Minimal bactericidal concentration (MBC) was recorded as the lowest concentration of the crude plant extract that killed 100% of the test organisms. MBC was determined by adding 50 µl aliquots of the serial dilution which did not show any visible growth after incubation in the MIC assay, and 50 µl of INT, to 100 µl of Muller-Hinton broth. The plates were incubated at 37℃ for 24 hours. The presence of bacterial growth was indicated by a pink/red colour, and clear wells indicated the inhibition of bacterial growth by the plant extract. The lowest concentration indicating inhibition of growth was recorded as the MBC.

### 2.6 Statistical analysis

The assay was performed in quadruplicate and repeated three times. All data was expressed as mean and standard deviation using MS Excel 2013 and ANOVA GraphPad Prism 5.

## 3.0 RESULTS

Antimicrobial effects of nine plant extracts were evaluated in terms of their ability to inhibit growth of the selected bacterial strains. Nine extracts were screened and found to be active against one or more of bacterial strains used (**see Table 2**). There was no significant difference observed between the solvents used in reconstitution of the extracts. For the results interpretation, an inhibition effect below 1 mg/ml was considered to be a good activity effect and anything above 1 was considered as moderate activity. Essentially above 2 mg/ml was not considered active effect in this study.

**Table 2**: Minimal inhibitory concentration of nine plant extracts expressed in mean and standard deviation.

| Plant Species | Plant part | Solvent | *E. coli* (mg/ml) | *Ps. aeruginosa* (mg/ml) | *S. aureus* (mg/ml) | *E. faecalis* (mg/ml) |
|---|---|---|---|---|---|---|
| | | | Mean± SDV | Mean± SDV | Mean± SDV | Mean± SDV |
| *Euclea crispa* | leaf | MeOH | 1.75±0.00 | 2.00±0.00 | 1.67±0.00 | 1.63±0.00 |
| | | DCM | 1.75±0.00 | 2.00±0.00 | 1.60±0.58 | 1.44±0.00 |
| | | EA | 1.28±0.00 | 1.50±0.71 | 1.34±0.00 | 1.38±0.00 |
| *Euclea natalensis* | leaf | MeOH | 1.25±0.00 | 1.00±0.00 | 1.33±0.00 | 1.12±0.00 |
| | | DCM | 1.75±0.00 | 2.00±0.00 | 1.60±0.58 | 1.55±0.58 |
| | | EA | 1.38±0.00 | 2.00±0.00 | 1.41±0.00 | 1.43±0.00 |
| *Schkuhria pinnata* | leaf | MeOH | 1.00±0.00 | 1.25±0.00 | 1.25±0.58 | 1.34±0.00 |
| | | DCM | 1.00±0.00 | 1.25±0.00 | 1.10±0.00 | 1.13±0.58 |
| | | EA | 0.50±0.00 | 0.63±0.00 | 0.74±0.58 | 0.94±0.00 |
| *Ziziphus mucronata* | leaf | MeOH | 0.50±0.00 | 0.63±0.00 | 0.82±0.58 | 0.83±0.00 |
| | | DCM | 1.00±0.00 | 0.88±0.00 | 0.84±0.00 | 0.82±0.00 |
| | | EA | 0.50±0.00 | 0.44±0.00 | 0.61±0.58 | 0.67±0.00 |
| *Ziziphus mucronata* | fruit | MeOH | 1.00±0.00 | 0.50±0.00 | 1.15±0.00 | 0.98±0.00 |
| | | DCM | 1.75±0.00 | 2.00±0.00 | 1.60±0.58 | 1.55±0.58 |

| | | | | | | |
|---|---|---|---|---|---|---|
| | | EA | 1.50±0.58 | 2.00±0.00 | 1.31±0.00 | 1.06±0.00 |
| *Lippia javanica* | leaf | MeOH | 0.25±0.00 | 0.50±0.87 | 0.53±0.00 | 0.49±0.00 |
| | | DCM | 0.50±0.00 | 1.13±0.29 | 1.11±0.58 | 1.03±0.29 |
| | | EA | 0.50±0.00 | 0.50±0.29 | 0.43±0.00 | 0.50±0.87 |

| Plant Species | Plant part | Solvent | E. coli (mg/ml) | Ps. aeruginosa (mg/ml) | S. aureus (mg/ml) | E. faecalis (mg/ml) |
|---|---|---|---|---|---|---|
| *Vernonia oligocephala* | leaf | MeOH | 1.06±0.87 | 1.00±0.00 | 1.26±0.00 | 1.26±0.58 |
| | | DCM | 1.25±0.00 | 1.00±0.00 | 1.25±0.58 | 1.34±0.00 |
| | | EA | 1.75±0.00 | 2.00±0.00 | 1.67±0.00 | 1.63±0.00 |
| *Clerodendrum myricoides* | leaf | MeOH | 1.75±0.00 | 2.00±0.00 | 1.52±0.58 | 1.49±0.58 |
| | | DCM | 1.25±0.00 | 1.00±0.00 | 1.17±0.58 | 1.17±0.58 |
| | | EA | 1.75±0.00 | 2.00±0.00 | 1.67±0.00 | 1.63±0.00 |
| *Erythrina lysistemon* | leaf | MeOH | 1.00±0.00 | 0.88±0.00 | 0.91±0.58 | 1.08±0.00 |
| | | DCM | 0.50±0.00 | 1.00±0.00 | 1.00±0.58 | 1.09±0.58 |
| | | EA | 0.50±0.00 | 0.44±0.00 | 0.50±1.01 | 0.52±0.00 |
| Gentamacin | | MeOH | 1.75±0.00 | 2.00±0.00 | 1.31±0.14 | 1.19±0.00 |
| | | DCM | 1.19±0.00 | 2.00±0.00 | 1.01±0.29 | 0.92±0.29 |
| | | EA | 1.19±0.00 | 2.00±0.00 | 1.21±0.58 | 1.14±0.00 |

MeOH: methanol; DCM: dichloromethane; EA: ethanol; *E. coli*: *Escherichia coli*; *Ps. aeruginosa*: *Pseudomonas aeruginosa*; *S. aureus*: *Staphylococcus aureus*; *and E. faecalis: Enterococcus faecalis.*

**Determination of minimal inhibitory concentration (MIC)**

Table two lists the antimicrobial activity of all plant extracts. The following four plant extracts showed good antimicrobial activity against all four bacteria used which are *Lippia javanica* (0.25±0.00 to 1.13±0.29mg/ml); *Ziziphus mucronata* leaf (0.44±0.00 to 1.00±0.00 mg/ml); *Erythrina lysistemon* (0.44±0.00 to 1.08±0.00 mg/ml) and *Schkuhria pinnata* (0.5±0.00 to 1.34±0.00 mg/ml). Furthermore, it can be seen that the other five plant extracts showed good inhibition effects against three bacterial straight which were below 1.75±0.00 mg/ml, except for *Ps aeruginosa* (2.00±0.00 mg/ml). This suggests *Ps aeruginosa* is resistance against plant extracts used. Similarly, gentamycin (positive control) had inhibitory effects ranged from 0.92±0.29 to 2.00±0.00 mg/ml against all four bacteria used.

**Determination of minimal bactericidal concentration (MBC)**

Table 3 represents minimum bactericidal effects of nine plant extracts against all four bacteria used. Interestingly, the four plant extracts that showed inhibition effects (**see Table 2**) have demonstrated bactericidal effects against three bacteria used (*E.coli*, *S. aureus* and *E. faecalis*). The plant extracts are *Lippa javanica* (0.25±0.00 to 1.00±0.00 mg/ml); *Ziziphus mucronata* leaf (0.25±0.00 to 2.00±0.00 mg/ml); *Erythrina lysistemon* (0.5±0.00 to 2.00±0.00 mg/ml) and *Schkuhria pinnata* (0.5±0.00 to 2.00±0.00 mg/ml. Moreover, the bactericidal effects for other four plant extracts was observed to be 1.67±0.58 against *E.coli*, *S. aureus* and *E. faecalis*. Essentially, no bactericidal effect were observed for *Euclea crispa*. None of the plant extracts used showed bactericidal effects against *Ps aeruginosa*. This indicates that *Ps aeruginosa* is residence against plant extracts tested against.

**Table 3**: Minimal bactericidal concentration of nine plant extracts expressed in mean and standard deviation.

| Plant Species | Plant part | Solvent | *E. coli* (mg/ml) | *Ps. aeruginosa* (mg/ml) | *S. aureus* (mg/ml) | *E. faecalis* (mg/ml) |
|---|---|---|---|---|---|---|
| | | | Mean± SDV | Mean± SDV | Mean± SDV | Mean± SDV |
| *Euclea crispa* | leaf | MeOH | 2.00±0.00 | 2.00±0.00 | 2.00±0.00 | 2.00±0.00 |
| | | DCM | 2.00±0.00 | 2.00±0.00 | 1.67±0.58 | 1.00±0.00 |
| | | EA | 1.00±0.00 | 2.00±0.00 | 2.00±0.00 | 2.00±0.00 |
| *Euclea natalensis* | leaf | MeOH | 1.00±0.00 | 2.00±0.00 | 2.00±0.00 | 0.50±0.00 |
| | | DCM | 2.00±0.00 | 2.00±0.00 | 1.67±0.58 | 1.00±0.00 |
| | | EA | 1.00±0.00 | 2.00±0.00 | 2.00±0.00 | 2.00±0.00 |
| *Schkuhria pinnata* | leaf | MeOH | 1.00±0.00 | 2.00±0.00 | 0.25±0.00 | 2.00±0.00 |
| | | DCM | 0.50±0.00 | 2.00±0.00 | 1.00±0.00 | 1.67±0.58 |

| Plant Species | Plant part | Solvent | *E. coli* (mg/ml) | *Ps. aeruginosa* (mg/ml) | *S. aureus* (mg/ml) | *E. faecalis* (mg/ml) |
|---|---|---|---|---|---|---|
| | | EA | 2.00±0.00 | 1.00±0.00 | 1.00±0.00 | 2.00±0.00 |
| *Ziziphus mucronata* | leaf | MeOH | 0.50±0.00 | 1.00±0.00 | 0.25±0.00 | 1.00±0.00 |
| | | DCM | 2.00±0.00 | 2.00±0.00 | 1.00±0.00 | 1.00±0.00 |
| | | EA | 0.50±0.00 | 1.00±0.00 | 1.00±0.00 | 0.50±0.00 |
| *Ziziphus mucronata* | fruit | MeOH | 0.50±0.00 | 2.00±0.00 | 2.00±0.00 | 0.50±0.00 |
| | | DCM | 2.00±0.00 | 2.00±0.00 | 1.67±0.58 | 1.67±0.58 |
| | | EA | 0.50±0.00 | 1.00±0.00 | 2.00±0.00 | 2.00±0.00 |
| *Lippia javanica* | leaf | MeOH | 0.25±0.00 | 0.50±0.00 | 0.25±0.00 | 0.50±0.00 |
| | | DCM | 1.00±0.00 | 2.00±0.00 | 0.71±0.51 | 1.00±0.00 |
| | | EA | 1.00±0.00 | 0.25±0.00 | 0.75±1.08 | 0.50±0.00 |
| *Vernonia oligocephala* | leaf | MeOH | 1.00±0.00 | 2.00±0.00 | 1.67±0.58 | 2.00±0.00 |
| | | DCM | 1.00±0.00 | 0.50±0.00 | 1.33±0.58 | 1.00±0.00 |
| | | EA | 2.00±0.00 | 2.00±0.00 | 1.67±0.58 | 2.00±0.00 |
| *Clerodendrum myricoides* | leaf | MeOH | 2.00±0.00 | 2.00±0.00 | 1.00±0.00 | 2.00±0.00 |
| | | DCM | 1.00±0.00 | 2.00±0.00 | 1.50±0.87 | 1.67±0.58 |
| | | EA | 2.00±0.00 | 0.50±0.00 | 0.42±0.14 | 2.00±0.00 |
| *Erythrina lysistemon* | leaf | MeOH | 1.00±0.00 | 1.00±0.00 | 0.50±0.43 | 2.00±0.00 |
| | | DCM | 2.00±0.00 | 2.00±0.00 | 1.42±1.01 | 0.67±0.29 |
| | | EA | 1.00±0.00 | 0.25±0.00 | 0.19±0.11 | 0.50±0.00 |
| Gentamacin | | MeOH | 2.00±0.00 | 2.00±0.00 | 0.25±0.33 | 1.00±0.00 |
| | | DCM | 0.06±0.00 | 2.00±0.00 | 0.83±1.01 | 0.50±0.43 |
| | | EA | 0.06±0.00 | 0.25±0.00 | 2.00±0.00 | 0.50±0.00 |

MeOH: methanol; DCM: dichloromethane; EA: ethanol; *E. coli*: *Escherichia coli*; *Ps. aeruginosa*: *Pseudomonas aeruginosa*; *S. aureus*: *Staphylococcus aureus*; *and E. faecalis: Enterococcus faecalis.*

## 4.0 DISCUSSION

The plant crude extracts were screened for their minimum inhibitory concentration and minimum bactericidal concentration using eight different concentrations; against four pathogenic organism. The findings of this research have shown that all plant extracts demonstrated inhibitory effects against pathogenic organisms tested against. However, only four plant extracts showed good inhibition effects against all three bacteria sued which are *Lippia javanica*, *Ziziphus mucronata leaf*, *Erythrina lysistemon* and *Schkuhria pinnata*. Similarly, positive control had inhibitory effects were comparable to those plant extracts gave good inhibition effects in all four pathogenic organisms used.

For the bactericidal effects, the four plant extracts that demonstrated good inhibition effects which are mentioned above also demonstrated good bactericidal effects only against three bacteria used. It was observed that *Ps. aeruginosa* did not demonstrate bactericidal effect against plant extract used.

The antimicrobial effects of plant extracts that showed inhibitory effects in our experiments are in agreement with their known medicinal use. For example, *Lippia javanica* has been reported to be used by Xhosa people to disinfect meat that has been contaminated by anthrax. In addition to that, it has been reported to be used for fever, cough, bronchitis and influenza (Van Wyk *et al*., 2009; Van Wyk, 2011). *Ziziphus mucronata* has been reported to have various medicinal used per each region and the most common one is to treat boils, swollen glands, wounds and sores (Palmer and Pitman 1972). Interestingly, *Erythrina lysistemon* bark is used to treat sores, wounds, abscesses and arthritis. Furthermore, the leaves infusions are used as ear drops to relieve earache, and decoctions of the roots are applied to sprains (Van Wyk *et al*., 1997).
Lastly, *Schuhria pinnate* have been used as a bactericide in open wounds, to treat acne, malaria and inflammation, and as a blood purifier and diuretic (Bussmann *et al*., 2008).

In this study, four microorganisms were used as to assess the ability of plant extracts to inhibit their growth. These were gram-negative [*E. coli* and *Ps. aeruginosa*] and gram-positive [*S.aureus* and *E. faecalis*]. It was observed that plant extracts demonstrated a degree of inhibition towards both gram-negative and gram-positive bacteria. Generally, gram-negative bacteria

are resistance to most of antibiotics, mainly lipophilic and amphiphilic. This is mainly due to outer membrane in gram negative bacteria, which excludes certain drugs and antibiotics from penetrating the cell (Miller, 2016; Mia-Prochnow, 2016). In contracts to gram positive bacteria do not have outer membrane, so they are more susceptible to antibiotics (Verma, 2012; Mai-Prochnow, 2016).

In our findings *Ps. aeruginosa* was observed to be resistance towards most plant extracts tested. This was has been previously reported in literature that *Ps. aeruginosa* has the ability to develop resistance to antibiotics rather rapidly over several generations. Essentially, resistance strains makes it difficult to treat once in a host, such as a human or other animal, is infected (Griffin *et al*., 2004). The sensitive organisms (*E.coli*) demonstrated to be sensitive in our findings and this is due to the fact that *E. coli* is well characterised, widely available and sensitive to antibiotics (Livermore *et al*., 2008).

## 5.0 CONCLUSION

To this effect, the findings suggest that there is a great need for further development of antimicrobial agents that may contribute to the improvement of future chemotherapeutic agents from medicinal plants. It was seen that *Lippia javanica*, *Ziziphus mucronata* leaf, *Erythrina lysistemon* and *Schkuhria pinnata* showed a good inhibitory and bactericidal effects against bacteria used. In four pathogenic organisms used, only *Ps. aeruginosa* demonstrated some degree on resistance is certain plant extracts used. Interestingly, *E.coli* was observed to be sensitive strain in almost all plant extracts tested followed by *S. aureus*. The plants contain diverse class of compounds which possess biologically active agents. The plant extracts have exhibited excellent antimicrobial activities, with minimum inhibitory concentrations comparable to those of standard drugs. The future work involves identification of the active compounds and characterization.

## 6.0 CONFLICT OF INTERESTS

The authors declare that they do not have any conflict with respect to the publication of this paper.

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