Synergistic Potential of the Leaves of Aspillia Africana (Compositae) and Psidium guajava (Myrtaceae) Against Some Selected Bacterial and Fungal Isolates

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Abstract

Background: The plethora of secondary metabolites inherent in plants that could be synthesized and formulated into useful drugs for the treatment of diseases is not fully understood.

Objectives: This research was carried out to investigate the antimicrobial potential of the leaves of Aspillia africana and Psidium guajava plants on some selected bacterial and fungal isolates.

Methods: Fresh and matured leaves of Aspillia africana and Psidium guajava were collected at the Federal College of Agriculture, Akure and the Botanical Garden, Obafemi Awolowo University (OAU), Ile-Ife, Nigeria. The plants were identified and voucher specimen deposited at IFE Herbarium. The pure isolates of the bacteria and fungi were obtained from Medical Microbiology unit, Department of Microbiology, Obafemi Awolowo University, Ile-Ife, Nigeria. The bacterial isolates were maintained on nutrient agar and the fungal isolates on Sabouraud Dextrose Agar (SDA). Extraction of the plant materials using methanol, ethanol and aqueous fractions were carried out using standard protocols. The isolates were inoculated on a nutrient broth. The Minimum Inhibitory Concentration (MIC) of the extracts was determined by broth dilution method while minimal bactericidal and fungicidal concentrations respectively were determined following established protocols.

Results and conclusion: The results showed that Bacillus cereus, Cornybacterium pyogenes, Klebsiella pneumonia and Escherichia coli showed higher degree of resistance to the plant extracts. The fungal isolates; Candida albicans, Trypophyton rubrum, Penicillium expansium, and Aspergillus flavus were resistant to Psidium guajava extract as no significant biological activity was observed. All the extracts from the plants produced considerable antimicrobial activities with Streptococcus faecalis and Staphylococcus aureus, whereas, these organisms were resistant to ofloxacin, sparifloxacin, chloramphenicol, augmentin, ciprofloxacin and septrin. This study concludes that ethanolic extracts (100mg/ml) of the leaf of Aspillia africana is more potent than the methanolic and aqueous extracts of Psidium guajava. This study demonstrated that the therapeutic properties of Aspillia africana and Psidium guajava are better enhanced with their synergistic potential to the tested microorganisms.

Keywords: Aspillia Africana, Bactericidal, Psidium guajava, Synergistic, therapeutic

INTRODUCTION

Crude extracts of medicinal plants notwithstanding stand out as veritable sources of potential resistance agents and the African biotopes promises to be a potential source of such compounds owing to its rich plant species diversity. According to World Health Organization, a larger percentage of the world’s population is based mainly on traditional medicine and a significant part of the therapy involves the use of plant materials or their bioactive components. There are many...
findings on the therapeutic potentials of plant extracts against bacteria\(^3\), hence, plants are still being recognized as the pivot of modern medicine for the prophylaxis of infectious diseases\(^4\). Therefore, the contributions of medicinal and pharmaceutically important plants to health care cannot be over-emphasized. There is a growing demand to discourage the usage of drugs with high clinical toxicity. However, the usual practice of oral administration of crude extracts poses threat to human health as the extracts may contain some toxic components\(^5\). These ambivalent challenges had necessitated the search for antibiotics in institutions, industries and the academic community \(^6\). Plants have major advantage over synthetic drugs because it’s cost effective, readily available, cheaper alternative and pharmacological importance \(^7\). Several studies have been reported on the antibacterial activities of local plants such as Euphorbia hirta, Kigelia Africana, Hibiscus sabdarifa and Mucuna pruriens\(^8\).

Aspillia africana (Compositae) is a tropical shrub, semi-woody herb from a perennial root widely grown in Nigeria where it is commonly known as Yunninyu by the Yorubas, Oraangila by Igbos and Tozalin by Hausas. The plant is about 2m high, polymorphic and ubiquitous on western land of Savannah. Its formulations had been used in the treatment of various bacterial infections such as gastro-intestinal disorders, corneal opacity and insect bites. The plant is widely used as haemostatic agent (spontaneous arrest of bleeding from damaged blood vessels). The fresh leaves are effective in wound healing, cuts and sores\(^9\).

| Table 1. Antimicrobial effect of methanolic (M), Ethanolic (E) and Aqueous (A) extracts of Aspillia africana and Psidium guajava on some selected bacterial and fungal isolates |
|---------------------------------|-----------------|-----------------|-----------------|
|                                | Aspillia africana | Psidium guajava |
|                                | (100mg/ml)       | (100mg/ml)      |
| Test bacteria                  | A    | E   | M   | A    | E   | M   |
| Providencia stuartii           | 12   | 4   |     | 8    | 12  | 4   |
| Bacillus cereus                | -    | -   |     | -    | -   | -   |
| Staphylococcus aureus          | 15   | 16  | 3   | 7    | 12  | 15  |
| Corynebacterium pyogenes       | -    | -   |     | -    | -   | -   |
| Streptococcus faecalis         | 4    | 8   | -   | 10   | 15  | 4   |
| Klebsiella oxytoca             | -    | -   |     | -    | -   | -   |
| Klebsiella pneumonia           | -    | -   |     | -    | -   | -   |
| Escherichia coli               | -    | -   |     | -    | -   | -   |
| Pseudomonas fluorescens        | 8    | 10  | -   | -    | -   | -   |
| Serratia rubidae               | -    | -   |     | 4    | 7   | -   |
| Proteus mirabilis              | -    | -   |     | -    | -   | -   |
| Salmonella pullorum            | -    | -   |     | -    | -   | -   |
| Test fungi                     | -    | -   |     | -    | -   | -   |
| Trychophyton tonsurans         | 1    | 2   | -   | -    | -   | -   |
| Candida albicans               | -    | -   | 1   | -    | -   | -   |
| Trychophyton rubrum            | -    | -   |     | -    | -   | -   |
| Penicillium expansum           | -    | -   |     | -    | -   | -   |
| Alternaria sp.                 | -    | -   |     | -    | -   | -   |
| Fusarium sp.                   | 3    | 5   | 2   | -    | -   | -   |
| Aspergillus niger              | -    | -   |     | -    | -   | -   |
| Aspergillus fumigates          | -    | -   |     | -    | -   | -   |
| Aspergillus flavus             | -    | -   |     | -    | -   | -   |
| Penicillium camemberti         | -    | -   |     | -    | -   | -   |

CLSI. Performance standards for antimicrobial susceptibility testing. Clinical and laboratory standards institute, 29\(^{th}\) ed, 2019.

Psidium guajava L. (Mytaceae) is one of such plants in folk medicine that had been extensively used for the treatment of various disease conditions like fever, dysentery, vomiting and inflamed gums\(^10\). The extract from the leaves were found to display analgesic and anti-inflammatory potential\(^11\), antimicrobial and antioxidant activities\(^12\). The alarming increase of resistance to antibiotics by microorganisms cannot be overemphasized. This had necessitated scientist to search for alternative strategies of mitigating this challenge. The objectives of the study are therefore to investigate the antimicrobial potential of the leaves of Aspillia africana and Psidium guajava plants on some selected bacterial and fungal isolates; and to investigate the synergistic antimicrobial effects of the extracts on the test organisms.

Materials and methods

Preparation and authentication of plant samples

A very fresh and matured leaves of Aspillia africana and Psidium guajava were collected at the Federal College of Agriculture, Akure and Botanical Garden, Obafemi Awolowo University (OAU), Ile-Ife, Nigeria. The specimens were identified at IFE Herbarium,
Department of Botany, Obafemi Awolowo University, Ile-Ife and voucher deposited. Each plant sample was separately washed and rinsed several times in distilled water. The samples were dried to constant weight in an oven and ground into powdery form using laboratory blender. The powder (100g) obtained were extracted with 500ml of sterile distilled water, 500ml ethanol and 500ml methanol by soaking in water for 5days and then filtered. The filtrates were later concentrated at 45°C using a motorized rotary evaporator (Resona England). The dried samples were kept in sterile bottles for future use.

Table 2. Synergistic antibacterial and antifungal effects of the plant extracts on the tested microorganisms

| Test bacteria            | A   | E | M (100mg/ml) |
|-------------------------|-----|---|--------------|
| Aspergillus africana    | 6   | 18| 15           |
| Psidium guajava          |     |   |              |
| Test fungi              |     |   |              |
| Trychophyton tonsurans  | -   | - | -            |
| Candida albicans         | -   | - | -            |
| Penicillium expansum     | -   | - | -            |
| Alternaria sp.           | -   | - | -            |
| Fusarium sp.            | 2   | 4 | 2            |
| Aspergillus niger        | -   | - | -            |
| Aspergillus fumigatus    | -   | - | -            |
| Aspergillus flavus       | -   | - | -            |
| Penicillium camembertii  | -   | - | -            |

KEY: A – Aqueous extracts, E – Ethanolic extracts, M – Methanolic extract

Preparation of the medium

40g of Mueller-Hinton agar powder were carefully weighed into a clean conical Flask and 100 ml of sterilized distilled water was dispensed into the conical flask to achieve homogeneity using magnetic stirrer before autoclaving at 121°C for 15 min. The plates were allowed to cool for 2h before a cork borer (No 4) was flamed and used to bore wells (6mm). The plant extracts were filled into the wells and labelled appropriately.

Preparation and Extraction of Plant Materials

Freshly harvested sampled leaves of A. africana and P. guajava were thoroughly washed with distilled and rinsed accordingly. Twenty gram each of the leaves was weighed on an electric weighing balance for methanol, ethanol and aqueous extraction. The leaves (20g) were later grounded with mortar and pestle and then transferred into labeled petridishes. Incubation was carried out at 37°C for 24h. About 90ml each of the solvent were added to the grounded leaves for extraction, vigorously shaken for 30 minutes and allowed to settle.

Sensitivity Test: Antibacterial and Antifungal assay was carried out following established protocols.

Minimal inhibitory concentration (MIC)

The minimum inhibitory concentration (MIC) of the extracts was determined following established protocols. About100mg/ml of the extracts were prepared and introduced into the test tubes containing 1ml of sterile Mueller-Hinton broth, 1ml of the different extract concentration and 1ml broth culture. The mixture was homogenized and then incubated at 37°C for 24h. The
estimation of minimal inhibitory concentration of the plant extracts on the fungi was determined using established protocols. Plant extracts was prepared and 2ml of different concentrations of the solution were added to Sabouraud Dextrose Agar (SDA) at 40°C to give the appropriate concentration. The medium was then poured to sterile Petri-dishes and allowed to cool. The extracts were then streaked with fungal spores. The plates were incubated at 25°C for 5days and were examined for the presence or absence of fungal growth. The MIC was taking as the lowest concentration that inhibited the growth of the test fungi.

**Minimal bactericidal and fungicidal concentration (MBC and MFC)**

Minimal bactericidal concentration (MBC) was determined by plating 1ml of the MIC positive tubes on nutrient agar to ascertain the bactericidal effect of the extracts. Samples were taken from plates with no significant growth in the MIC culture and sub-cultured on freshly prepared Mueller-Hinton agar plates for determination of MBC and SDA plates for MFC and later incubated at 25°C for 5days.

**Synergistic antimicrobial effects of the plant extracts against tested bacterial and fungal isolates:**

The synergistic antimicrobial effects of the various combinations of the plant extracts for the test microorganisms were determined using the method employed by. To demonstrate synergism, equal quantity of plant combinations were weighed and dissolved in the appropriate quantity of the dilution to give the 100mg/ml concentrations which was used for the antimicrobial test. Antimicrobial susceptibility testing will be compared with clinical and laboratory standard institute (CLSI, 2019).

**Results**

**Antimicrobial effect of methanolic (M), Ethanolic (E) and Aqueous (A) extracts of* Aspillia africana* and* Psidium guajava* on some selected bacterial and fungal isolates:**

The zones of inhibition of the extracts ranged from 1-15mm for bacteria while fungal isolates ranged between 1 and 7mm respectively. The highest and lowest zones of inhibition of 15mm and 3mm respectively were obtained in *Staphylococcus aureus* under the aqueous and methanolic fractions. *Bacillus cereus, Corynbacterium pyogenes, Klebsiella pneumonia* and *Escherichia coli* showed high degree of resistance to the plant extracts. The fungal isolates; *Candida albicans, Trypophyton rubrum, Penicillium expansium,* and *Aspergillus flavus* were resistant to *Psidium guajava* extract as no significant activity was observed. There were no significant effect of the plant extracts against tested fungi isolates (Table 1).

**Synergistic antibacterial and antifungal effects of the plant extracts on the tested microorganisms:**

The fungal isolates were resistant to the plant extracts as no significant effect was observed. However, the highest zone of inhibition of 21mm was observed in S. aureus under the ethanolic fraction while the lowest (1.2mm) was noticeable in *Serratia rubidae* under the methanolic extract (Table 2). *Escherichia coli* and *Klebsiella pneumonia* were both resistant to the synergistic effect of the plant extracts.

**Susceptibility of the test organisms to reference drugs**

The susceptibility of the test organisms to antibiotics and antifungal drug is presented in Table 3. Some of the microorganisms which were sensitive to the plant extracts were found to be resistant to some of the antibiotics used. All the plant extracts produced considerable antimicrobial activities with *Strepotococcus faecalis* and *S. aureus*, whereas, these organisms were resistant to ofloxacin, sparflxacin, chloramphenicol, augmentin, ciproflxacin and spetrin. However, the antifungal drug (Ketoconazole) was more effective on the test fungi than the plant extracts.

**Minimum inhibitory concentrations (MICs) of the plant extracts against the tested microorganisms**

The MIC ranged from 25 to 250µg/ml. The lowest MIC (25µg/ml) was obtained against *P. guajava* and *A. africana* with ethanolic extract of *P. guajava* and had a MIC of 25µg/ml on *S. aureus*. The highest MIC (250µg/ml) was obtained with the aqueous extract of *A. africana* with *P. stuartii* against *P. fluorescens* and methanolic extract of *P. guajava* on *S. faecalis* (Table 4). The MIC of the plant extracts against selected fungi ranged from 150-300µg/ml. Aqueous and ethanolic extracts of *A. africana* recorded MIC of 250 µg/ml on *T. tonsurans* and *Fusarium* sp. respectively. The highest MIC (300 µg/ml) was obtained with methanolic extract of *A. africana* on *Fusarium* sp (Table 4).

**Minimum bactericidal concentrations (MBCs) of the plant extracts against the tested microorganisms**

The MBC ranged from 50-400µg/ml. The lowest MBC (50µg/ml) was obtained with ethanolic and aqueous extract of *P. guajava* against *S. aureus*. The highest MBC (400µg/ml) was obtained with the aqueous and ethanolic extract of *A. africana*, and methanolic extract of *P. guajava* on *S. faecalis* respectively (Table 5).

**Discussion**

Nigeria is repleted with reservoir of medicinal plants. The economic burden and health implications of antimicrobial resistance are quite alarming. The unquantifiable extent of the antimicrobial potency of plant extracts orchestrated by characterization of abundant secondary metabolites inherent in these plants necessitated research on traditional medicine. Findings from this study revealed that aqueous and ethanolic extracts of the leaf of *Aspillia africana* is more potent than the ethanolic extract of *Psidium guajava*. *A. Africana* possess antibacterial activity against *Staphylococcus aureus*, *Strepotococcus faecalis* and *Pseudomonas fluorescens*. reported earlier that the ethanolic extract of the leaf of *A. africana* possess antibacterial potential against these microorganisms. However, *Psidium guajava* exhibited no antimicrobial activity against the tested fungal isolates. Among the bacterial isolates *Staphylococcus aureus* were the most sensitive to the plant extracts while
### Table 3. Susceptibility of the test organisms to reference antibiotics and antifungal drugs

| Test Bacteria                        | Diameter of Zones of Inhibition (µg) |
|--------------------------------------|--------------------------------------|
|                                      | OFL (5)     | SP (10) | CHL (30) | AUG (5) | CPX (5) | SXP (25) | AMX (10) | GEN (5) | PEF (5) | STR (10) | APX (10) | Z (30) | CT (10) | ERY (10) | N (10) | CM (10) |
| Gram negative                        |            |         |         |         |         |         |         |         |         |         |         |         |         |         |         |         |
| Escherichia coli                     | 15 (R)     | 17 (R)  | 14 (R)  | 13 (R)  | 18 (S)  | 13 (R)  | 13 (R)  | 16 (S)  | 17 (S)  | 14 (R)  | -        | -        | -        | -        | -        | -        |
| Klebsiella pneumoniae                | 0 (R)      | 0 (R)   | 15 (S)  | 20 (S)  | 0 (R)   | 12 (R)  | 16 (R)  | 12 (R)  | 23 (S)  | 0 (R)   | -        | -        | -        | -        | -        | -        |
| Klebsiella oxytoca                   | 0 (R)      | 16 (R)  | 20 (S)  | 16 (R)  | 18 (S)  | 22 (S)  | 16 (R)  | 14 (R)  | 20 (S)  | 18 (S)  | -        | -        | -        | -        | -        | -        |
| Pseudomonas fluorescens              | 0 (R)      | 13 (R)  | 20 (S)  | 12 (R)  | 18 (S)  | 14 (R)  | 20 (R)  | 16 (S)  | 12 (R)  | 20 (S)  | -        | -        | -        | -        | -        | -        |
| Serratia rubidae                     | 4 (R)      | 4 (R)   | 5 (R)   | 0 (R)   | 12 (R)  | 0 (R)   | 0 (R)   | 8 (R)   | 16 (S)  | 12 (R)  | -        | -        | -        | -        | -        | -        |
| Proteus mirabilis                    | 8 (R)      | 6 (R)   | 2 (R)   | 0 (R)   | 16 (R)  | 20 (S)  | 4 (R)   | 22 (S)  | 16 (S)  | 2 (R)   | -        | -        | -        | -        | -        | -        |
| Salmonella pullorum                  | 19 (S)     | 0 (R)   | 2 (R)   | 0 (R)   | 0 (R)   | 16 (S)  | 0 (R)   | 21 (S)  | 22 (S)  | 0 (R)   | -        | -        | -        | -        | -        | -        |
| Providential stuartii                | 4 (R)      | 4 (R)   | 5 (R)   | 0 (R)   | 12 (R)  | 0 (R)   | 0 (R)   | 8 (R)   | 16 (S)  | 12 (R)  | -        | -        | -        | -        | -        | -        |
| Bacillus cereus                      | -          | -       | -       | -       | -       | -       | 0 (R)   | 24 (S)  | 26 (S)  | 20 (S)  | 0 (R)   | 24 (S)  | 0 (R)   | 4 (R)   | 8 (R)   | 10 (R)  |
| Staphylococcus aureus                | -          | -       | -       | -       | -       | -       | 0 (R)   | 22 (S)  | 18 (S)  | 0 (R)   | 0 (R)   | 0 (R)   | 0 (R)   | 19 (S)  | 0 (R)   |         |
| Corynebacterium pyogenes             | -          | -       | -       | -       | -       | -       | 12 (R)  | 3 (R)   | 21 (S)  | 0 (R)   | 19 (S)  | 12 (R)  | 4 (R)   | 10 (R)  | 19 (S)  | 0 (R)   |
| Streptococcus faecalis               | -          | -       | -       | -       | -       | -       | 12 (R)  | 17 (S)  | 7 (R)   | 0 (R)   | 8 (R)   | 10 (R)  | 10 (R)  | 15 (S)  | 6 (R)   | 0 (R)   |

| Test Fungi                           | Concentrations of Ketofung (mg/ml) |
|--------------------------------------|-------------------------------------|
|                                      | 05  | 10  | 20  |
| Trychophyton tonsurans               | 18  | 34  | 37  |
| Candida albicans                     | 18  | 25  | 33  |
| Penicillium expansum                 | 16  | 22  | 28  |
| Alternaria sp.                       | 11  | 13  | 17  |
| Fusarium sp.                         | 21  | 26  | 32  |
| Aspergillus niger                    | 16  | 18  | 24  |
| Aspergillus fumigatus                | 18  | 25  | 30  |
| Aspergillus flavus                   | 20  | 22  | 28  |
| Penicillium camemberti               | 14  | 18  | 22  |
| Trychophyton rubrum                  | 17  | 22  | 27  |

OFL= oxofloxacin (5µg) , SP= Sparfloxacin (5µg) , CHL= Chloramphenicol (30µg) , AUG= Augmentin (30µg), CPX= Ciprofloxacin (5µg), SXP= Seprtin (25µg), AMX= Amoxicillin (25µg), GEN= Gentamycin (10µg), PEF= Pefloxacin (5µg), STR= Streptomycin (10µg), APX= Ampiclox (10µg), Z= Zinnacef (µg), CT= Ceftriazone (30µg), ERY= Erythromycin (10µg), N= Nitrofurantoin (µg), CM= Clarithromycin (µg)
Corynebacterium pyogenes and Escherichia coli were the most resistant. The prolific synergistic sensitivity of the test isolates to aqueous and ethanolic extracts of both plants is at variance with the findings of 17, who reported that ethanol as the best solvent for the extraction of bioactive components from plants. This study showed that A. africana extract was active on both Gram-negative (P. fluorescens and P. stuartii) and Gram-positive (S. aureus and S. faecalis) bacterial at 100mg/ml. Similar findings were reported by 18 on the evaluation of aqueous extracts of the leaves of A. Africana in Sierra Leone which exhibited antibacterial activities on both Gram-positive and Gram-negative organisms at concentrations which ranged from 0.1-0.5g/ml. However, the present study contradicts the findings of 17 who reported aqueous extract of A. africana to be active on E. coli and Salmonella sp at concentrations ranging from 0.1-0.5g/ml. In the present study, the extract of Ps. guajava exhibited inhibitory property on S. aureus, S. faecalis and P. Staurtii thus conforms to earlier reports by 19-20 and 21 who reported the antibacterial activity of Ps. guajava leaf. 20 also reported the efficacy of the plant extract in the treatment of diarrhoea, dysentery and wounds. Most of the microorganisms tested in this study have been implicated in various gastrointestinal infections such as watery diarrhoea, nausea, vomiting, abdominal pain, pediatric diarrhoea, typical gastroenteritis, necrotizing enterocolitis and/or dysentery (E. coli, Klebsiella sp. and Salmonella sp) and wound infection (Staphylococcus sp., Streptococcus sp., Proteus sp. and Pseudomonas sp). The synergistic antimicrobial effect of the various plants extracts showed that the zones of inhibition increased considerably compared to when extracts were used singly. The wider zones of inhibition recorded may be due to resultant effects of the bioactive components in the extracts. 21 reported in their studies on in vitro synergistic activities of some herbs on S. typhi, that antimicrobial activity of some plants extracts increase when used synergistically. However, E. coli, P. mirabilis, P. expansium, Alternaria spp, A. fumigatus, A. flavus and P. carmenbirti all displayed resistance to all the combined extracts tested. This may be due to the possibility that the active ingredient of one particular plant may not be complementary to another, thereby resulting in neutralization or denaturalisation of the plants components. The results obtained from the minimum inhibitory concentration (MIC) of the various plants extracts showed that the plant extracts are very potent against the pathogens at the MIC of 25µg/ml. The high MIC obtained on the test fungi confirmed their low susceptibility to virtually all the extracts tested. The MICs of the fungal isolates ranged between 150 and 300µg/ml. The low susceptibility of these fungi to these extracts may likely be attributed to the fact that majority of fungi interact with plants in the field and would have naturally developed resistance to the active antimicrobial ingredients in these plants.

CONCLUSION

This study concludes that ethanolic extracts (100mg/ml) of the leaf of Aspillia africana is more potent than the methanolic and aqueous extracts of Psidium guajava. The plant extracts showed greater antimicrobial activity on the bacterial isolates than on the fungal isolates suggesting a broader spectrum of activity with ethanolic extract on the gram positive and the gram negative bacteria. The therapeutic properties of Aspillia africana and Psidium guajava is better enhanced with their synergistic potential to the tested microorganisms.

| Test Bacteria       | Aspillia africana | Psidium guajava |
|---------------------|-------------------|-----------------|
| Providencia stuartii | - 50 M            | A E             |
| Bacillus cereus     | - - 50 50 25      |
| Staphylococcus aureus | 50 50 100 -       |
| Streptococcus faecalis | 200 200 100 25 25 |
| Klebsiella oxytoca  | - - 100 100       |
| Klebsiella pneumonia| - - 200 -         |
| Escherichia coli    | - - 200 -         |
| Pseudomonas fluorescence | 250 200 -        |
| Salmonella pullorum | - - 250 -         |
| Corynebacterium pyogenes | - - - -         |
| Serratia rubidae    | - - - -           |

Test Fungi

| Test Fungi     | Aspillia africana | Psidium guajava |
|----------------|-------------------|-----------------|
| Trychophyton tonsurans | 250 250          |
| Candida albicans       | 250 - - -        |
| Trychophyton rubrum    | - - - - -        |
| Fusarium H.            | 250 250 -        |
| Aspergillus niger      | - - 250 -        |

KEY: A – Aqueous extracts, E – Ethanol extracts, M – Methanolic extract

| Test Bacteria     | Aspillia africana | Psidium guajava |
|-------------------|-------------------|-----------------|
| Providencia stuartii | 150 150 100 50   |
| Bacillus cereus    | 300 - - -        |
| Staphylococcus aureus | 150 150 300 50 50 |
| Streptococcus faecalis | 400 400 250 200 |
| Klebsiella oxytoca | 200 - - -        |
| Klebsiella pneumonia| 400 - - -        |
| Escherichia coli   | - - - - -        |
| Pseudomonas fluorescence | 300 400 -        |
| Salmonella pullorum| - - - - -        |
| Corynebacterium pyogenes | - - - - -    |
| Serratia rubidae   | 200 250 -        |

RECOMMENDATION

A further study on the proper formulation into drugs of the plant extracts for human consumption is highly
recommended. The toxicological studies of the extracts on experimental animals should be determined in order to ascertain its safety threshold on humans.

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