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**EFFICACY OF 13 MEDICINAL PLANTS USED BY INDIGENOUS COMMUNITIES  
AROUND LAKE VICTORIA, KENYA, AGAINST TUBERCULOSIS, DIARRHOEA  
CAUSING BACTERIA AND CANDIDA ALBICANS**

**Richard M. Mariita<sup>1</sup>, Paul O. Okemo<sup>1\*</sup>, John A. Orodho<sup>2</sup>, Claude Kirimuhuzya<sup>3</sup>, Joseph N. Otieno<sup>4</sup>,  
Magadula J. Joseph<sup>4</sup>**

<sup>1</sup> Plant and Microbial Sciences Department Kenyatta University, P.O Box 43844-00100, Nairobi, **Kenya.**

<sup>2</sup> Educational Management, Policy and Curriculum Studies, Kenyatta University, P.O Box 43844-00100,  
Nairobi, **Kenya.**

<sup>3</sup> Departments of Botany & Pharmacology and Therapeutics, Makerere University, P.O. Box 7062, Kampala,  
**Uganda.**

<sup>4</sup> Institute of Traditional Medicine, Muhimbili University, P.O Box 65001, Dar es Salaam, **Tanzania.**

Email: [okemo1952@yahoo.com](mailto:okemo1952@yahoo.com)

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**Abstract**

**Aims:** To investigate Crude methanol extracts of 13 medicinal plants obtained through an ethnobotanical survey against 4 strains of mycobacteria (*Mycobacterium tuberculosis*, *M. kansasii*, *M. fortuitum* and *M. smegmatis*), *Salmonella typhi*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Klebsiella pneumonia* and *Candida albicans*.

**Materials and Methods:** Antimycobacterial susceptibility tests were carried out using BACTEC MGIT 960 system. Evaluation of antibacterial, antifungal and phytochemical properties was done using standard procedures.

**Results and Discussion:** All the plant extracts inhibited mycobacterial growth at 2.0 mg/mL. *Carissa edulis* and *Vernonia amygdalina* were the most potent against *M. smegmatis* and *M. fortuitum*, completely inhibiting their

growth (Zero GUs) at all concentrations used. *Toddalia asiatica* had high inhibitory activity (Zero GUs) against *M. tuberculosis* and *M. kansasii* at all concentrations used. There was a significant difference on general antibacterial results of the extracts at  $P \leq 0.05$  against other test cultures. The most potent antibacterial extract was from *Toddalia asiatica* with an MIC and MBC of 9.375 mg/mL. *Carissa edulis* and *Momordica charantia* both produced MICs and MBCs of 37.5 mg/mL against *S. typhi* and *S. aureus*. *Lantana camara* produced MICs and MBCs of 37.5 mg/mL against both *S. aureus* and *P. aeruginosa*. Preliminary phytochemistry identified six phytochemicals with flavonoids being found in all extracts.

**Conclusion:** The data suggests that methanolic extracts of some of the plant species can be used against several microbial agents. Further work on them is underway.

**Key words:** Antimicrobial, Diarrhoeal, Lake Victoria region, Medicinal plants, Tuberculosis, phytochemicals

## **INTRODUCTION**

Microorganisms are frequently a cause of prevailing diseases, presenting a serious public health issue in a significant segment of the population as showed by both private and official health care systems. Currently, about 80% of the populations in developing countries use traditional medicine for their healthcare (1) with modern pharmacopoeias containing at least 25% of drugs derived from plants and many others which are synthetic analogues build on prototype compounds isolated from plants.

Neglected diseases are infectious diseases that primarily, though not exclusively, affect vulnerable populations in developing countries where poor sanitation and lack of access to health care foster disease transmission and vector proliferation (2). These diseases, which include tuberculosis, diarrheal diseases, and typhoid, cause 35,000 deaths per day in the developing world along with significant morbidity.

*Mycobacterium tuberculosis* is the main causative agent of tuberculosis (TB), an illness responsible for 26% of all possibly preventable deaths in the world (3). The drawbacks of the current existing drugs including the emergence of multi drug-resistant (MDR) and extensively drug resistant (XDR) strains have led to a renewed interest in the discovery of new anti-tubercular agents with novel modes of actions (4). The worldwide emergence of *Escherichia coli*, *Klebsiella pneumoniae* and many other  $\beta$ -lactamase producers has become a major therapeutic problem. Multi-drug resistant strains of *E. coli* and *K. pneumoniae* are widely distributed in hospitals and are increasingly being isolated from community acquired infections (5). There is great interest therefore, in the public health community in developing new products to treat or prevent these diseases (2). Screening of plant extracts is a good starting point for antimicrobial drug discovery (4). Higher plants remain a vital source of these new substances, especially in low resource countries (6). Various studies have pointed out the expressive richness of plants in traditional pharmacopoeias (7). Researchers consider them a source of diverse chemical structures that are virtually impossible to replicate in a synthetic chemistry laboratory (4).

Since the discovery of the first antibiotic, penicillin, the need for antimicrobial agents is yet to be satisfied because of the emergence of pathogenic microbes with increased resistance to established antibiotics (1, 4). This has been fuelled by the economic crisis, high cost of industrialized medicines and inefficient public access to medical and pharmaceutical care. There are also side effects caused by synthetic drugs which add to additional factors contributing to the exigent need for novel antimicrobial remedies (6, 8, 9).

## **MATERIALS AND METHODS**

### **Plant material**

Thirteen plant materials were used in this study. Plants were collected based on information gathered from the ethnobotanical survey (Appendix 1) carried out between September to November 2007 around the Lake

Victoria region. The various locations around Lake Victoria region represent diverse cultural backgrounds. The information obtained included local names of plants, traditional usage of the plants which the plant parts used. Plant materials were authenticated by a taxonomist, and voucher specimens deposited at the Department of Pharmacy and Complimentary Alternative Medicine herbarium, Kenyatta University, Nairobi, Kenya.

### **Preparation of extracts**

Collected plant parts were chopped and shade dried, then ground into powder using hammer type milling machine (Meecon, CM/L-1364548, India). Plant powder was soaked in methanol and extracted using soxhlet extractor for 72 h (10). The extracts were filtered through a Whatmann filter paper No. 42 (125 mm) and concentrated using a rotary evaporator (Laborota 4000, SN 090816862, Germany) with the water bath set at 40°C (11), then dried in a dessicator over anhydrous CuSO<sub>4</sub>. The extracts were thereafter subjected to biological assays against selected mycobacteria, bacteria and *Candida albicans*.

### **Test cultures**

Mycobacteria test cultures used for the bioassays included *Mycobacterium tuberculosis*, *M. kansasii*, *M. smegmatis* and *M. fortuitum* and were obtained from the Center for Respiratory Diseases Research (CRDR), Kenya medical Research Institute (KEMRI), Nairobi, Kenya. Other test cultures that included *Salmonella typhi* (clinical isolate), *Klebsiella pneumoniae* (clinical isolate), *Pseudomonas aeruginosa* (ATCC 25852), *Escherichia coli* (ATCC 25922) *Staphylococcus aureus* (ATCC 20591) and *Candida albicans* (ATCC EK138), a yeast like fungi, were obtained from Kenyatta National Hospital in Nairobi, Kenya.

## **Susceptibility tests**

### **Antimycobacterial susceptibility testing using BACTEC MGIT 960 System**

The BACTEC MGIT 960 is a fully automated, high volume; nonradiometric instrument that offers continuous monitoring of culture growth (12). The BACTEC MGIT 960 was used in the antimycobacterial activity assessment of the plant extracts. The extracts were dissolved in 0.01% DMSO and diluted to final concentrations of 0.5, 1.0 and 2.0 mg ml<sup>-1</sup>. A stock solution of 2.0 mg ml<sup>-1</sup> of isoniazid was used as the positive and 0.01% DMSO as the negative controls, respectively. Extract concentrations of 1 mL were introduced into each of the 7-mL mycobacterial growth indicator (MGIT) culture tubes, which contained Middlebrook 7H9 broth base enriched with oleic acid, albumin, dextrose, and catalase (BBL MGIT OADC) and an antibiotic mixture of polymyxin B, amphotericin B, nalidixic acid, trimethoprim, and azlocillin (BBL MGIT PANTA). Inoculated tubes were incubated at 37 °C in the BACTEC MGIT 960 for up to 42 days (Maximum protocol length 56 days). The BACTEC MGIT 960 monitors specimens hourly for an increase in fluorescence, at which time; the operator is audibly and visibly alerted to the location of tubes sensing the presence of mycobacteria. All specimens identified as positive by MGIT were confirmed using subsequent AFB smears. If no AFB were observed, the MGIT tube was returned to the instrument and incubated further for a total of 42 days. Culture vials which remained negative for a minimum of 42 days (maximum protocol length of 56 days) were removed and recorded as negative, while growth units (GUs) for the positive ones were recorded appropriately (13).

### **Evaluation of Antibacterial and Antifungal activities**

#### **Zones of inhibition**

The antibacterial and antifungal activities of extracts from the 13 plant species were assayed *in vitro* by agar disc diffusion (DD) method (14). Mueller Hinton agar and Potato Dextrose Agar (PDA) were prepared using

manufactures' instructions for purposes of culturing the bacteria and fungi respectively. Normal saline solution was used to dilute a 24 h culture of the bacterial type culture or clinical isolate to attain a 0.5 McFarland standard. Spread plate method was used to culture 0.1 mL of the microbial suspension that was introduced into the Petri dishes (11, 14). Filter paper discs (6 mm) were impregnated with the plant extracts. Eighteen dry sterile discs (6 mm diameter) were soaked in the plant extract (made by dissolving 300 mg of the extracts in 1 mL of methanol) air dried and placed on the spread plates at reasonable distances. Discs impregnated with methanol and air dried were used as negative controls together with various standard conventional antibiotics (Amoxicillin (Hangzhou Ruijian chemical Co., Ltd., batch 490805241); Ciprofloxacin (Chengdu Ware Yuanheng Pharmaceutical Co., Ltd, batch 20070907); Fluconazole (Pfizer Ltd., UK batch 30) as positive controls. The plates were then incubated at 35 °C for 24 h. This was replicated three times for each pathogen.

*Candida albicans* was cultured by taking 0.1 mL from the broth and spreading on PDA. The culture was incubated at 25 °C for 72 h. The cork boarer was used to pick a section of the young mycelium which was placed at the centre of the PDA plate and the dry discs which were impregnated with 0.1 mL of the plant extracts placed at a distance around the inoculum mycelium. The inoculum was incubated at 25 °C for 72 h. Fluconazole and dry discs treated with methanol were also used as positive and negative controls respectively. All tests were performed in triplicate. Microbial growth inhibition was determined by measuring the zones of inhibition using a transparent ruler.

### **Evaluation of Minimum Inhibitory concentrations (MICs) and Minimum Fungicidal concentrations (MFCs)**

A broth micro-dilution technique was adopted using 96 well micro-titer plates. This was done only where the plant extract showed strong antibacterial activity by the disk diffusion method ( $\geq 9$ -15 mm) (15). The wells

were filled with 50 µl of the Nutrient broth for bacterial strains and Potato dextrose broth for *C. albicans*. The extract was then prepared by taking 300 mg of the plant extract and mixing it with 1 mL of DMF (0.01% Dimethyl formamide) for complete dissolution of the extract. Then 50 µl of the plant extract was dispensed into the first well before serial dilutions were done by transferring 50 µl of nutrient or potato dextrose broth containing the extract from the first well to the second well, and from the second well to the third well through the fourth well. Fifty microlitres (50 µl) of the test isolate was then dispensed into each well. One well (without extract or drug) was used as negative control of the growth of the microorganisms in the medium whereas another well with 50 µl of the antibiotic (Amoxicillin /Ciprofloxacin/fluconazole) was used as positive control. Incubation was done at 37 °C for 24 h. The MIC values were determined as the lowest concentrations of the extract capable of inhibiting microbial growth.

For the determination of MBC/MFC, all wells where there was no growth were subcultured on nutrient agar and PDA. The lowest concentration of the plant extracts that did not yield any colony on the solid medium (Nutrient or PDA agar) after sub culturing and incubating for 24 h for bacterial strains and 72 h for *C. albicans* was taken as the MFC/MBC. All tests were performed in triplicates.

### **Phytochemical analysis**

Chemical constituents of the extracts were analyzed using the methodology described by Aiyelaagbe and Osamudiamen, (10) and Edeoga *et al.*, (11). Each extract was screened for presence of alkaloids, flavonoids, cardiac glycosides, tannin saponins and terpenoids.

### **RESULTS AND DISCUSSION**

The present study aimed at evaluating the *in vitro* antimicrobial activity of 13 selected medicinal plants used by the native communities living around the Lake Victoria region against bacterial and fungal isolates.

## **Mycobacteria**

There was complete inhibition (Zero GUs) of mycobacterial test strains used by all the plant extracts at a concentration of 2 mg ml<sup>-1</sup> (Table 1). *Carissa edulis* and *V. amygdalina* completely inhibited (Zero GUS) the growth of the fast growing mycobacteria, at all concentrations. *Croton macrostachyus* gave appreciable inhibitions against the two fast growing mycobacteria (76 GUs for *M. fortuitum* and 48 GUs for *M. smegmatis*) compared to the negative control (893 and 16017 GUs respectively) and also gave appreciable results against *M. tuberculosis* (398 GUs compared to the negative control which had 18683 GUs), while completely inhibiting the growth of *M. kansasii* (Zero GUs). *Vernonia amygdalina* also completely inhibited (Zero GUs) the growth of *M. kansasii* and gave appreciable results against *M. tuberculosis* (451 GUs compared to the negative control which had 18683 GUs). The root extract of *Toddalia asiatica* completely inhibited the growth *M. tuberculosis* and *M. kansasii* at all concentrations. *Lantana camara* only gave appreciable results against *M. tuberculosis* (128 GUs compared to the negative control which had 18683 GUs). For an extract to be said to be active against mycobacteria, it should have 99% inhibitory effect (13).

The antimicrobial properties of medicinal plants have been explained by the chemical association of active substances (16). *Vernonia amygdalina* extract had flavonoids as the only phytochemicals present in high concentration (Table 2). Flavonoids in *V. amygdalina* are reported to exhibit antioxidant activity and are effective scavengers of superoxide anions (17). Thus this can significantly affect the cell wall of the test culture which invariably may lead to the collapse of the cell wall and overall, affect the entire mechanism of the organism as reported by Nwinyi *et al.*, (17).

From this study, *Toddalia asiatica* which was found to have high inhibitory activity against *M. kansasii* and *M. tuberculosis* (Zero GUs at 0.5mg/ml) was found to have moderate quantities of flavonoids, whereas tannins and alkaloids were found in trace amounts (Table 2). More than 6400 flavonoids have been shown to have various



interesting properties, including antibacterial, antiprotozoal, anti-inflammatory, dietary, antioxidant, vascular and oestrogenic activities, mainly mediated through inhibition of oxidases (18). Some have been found to inhibit *de novo* fatty acid biosynthesis in mycobacteria as well as mycolic acid biosynthesis (18).

Flavonoids and tannins, which were detected in high and moderate concentrations respectively in *Carissa edulis*, could be responsible for the inhibitory effect of the plant. Different mechanisms have been linked to flavonoid-mediated cytotoxicity, including proteasome inhibition, inhibition of fatty acid synthesis, topoisomerase inhibition, inhibition of phosphatidyl-inositol 3-kinase, induction of cell cycle arrests, accumulation of p53 or enhanced expression of c-fos and c-myc (19). The 4-carbonyl group of flavonoids has been reported to correlate with inhibition of fatty acids. The presence of the 2–3 double bonds has been linked to efficient binding and inhibition of the Pglycoprotein (P-gp). Cheng and Pieters (20) have reported that novel proteasome inhibitors could be used as potential drugs to combat tuberculosis.

**Table 1: Antimycobacterial activity (GUs) of medicinal plants using BACTEC MGIT™ 960 System**

| Botanical name of plant | Activity on slow growers at various concentrations (mg/ml) |     |       |       |      |      | Activity on fast growers at various concentrations (mg/ml) |    |     |      |      |       |
|-------------------------|--|-----|-------|-------|------|------|--|----|-----|------|------|-------|
|                         | 2  |     | 1     |       | 0.5  |      | 2  |    | 1   |      | 0.5  |       |
|                         | Mk   | Mtb | Mk    | Mtb   | Mk   | Mtb  | Mf   | Ms | Mf  | Ms   | Mf   | Ms    |
| <i>C. edulis</i>        | 0  | 0   | 0     | 206   | 770  | 294  | 0  | 0  | 0   | 0    | 0    | 0     |
| <i>C. macrostachyus</i> | 0  | 0   | 0     | 160   | 0    | 398  | 0  | 0  | 0   | 0    | 76   | 48    |
| <i>V. amygdalina</i>    | 0  | 0   | 0     | 67    | 0    | 451  | 0  | 0  | 0   | 0    | 0    | 0     |
| <i>M. charantia</i>     | 0  | 0   | 3013  | 21408 | ND   | ND   | 0  | 0  | 113 | 5012 | ND   | ND    |
| <i>O. gratissimum</i>   | 0  | 0   | 678   | 10990 | ND   | ND   | 0  | 0  | 56  | 371  | ND   | ND    |
| <i>L. camara</i>        | 0  | 0   | 5775  | 0     | 9198 | 128  | 0  | 0  | 37  | 75   | ND   | ND    |
| <i>C. myricoides</i>    | 0  | 0   | 0     | 2212  | 1974 | 6752 | 0  | 0  | 0   | 0    | 438  | 10561 |
| <i>T. asiatica</i>      | 0  | 0   | 0     | 0     | 0    | 0    | 0  | 0  | 1   | 0    | 1205 | 138   |
| <i>L. trifolia</i>      | 0  | 0   | 16648 | 6310  | ND   | ND   | 0  | 0  | 75  | 89   | ND   | ND    |

|                      |     |      |       |       |       |       |     |      |     |      |     |       |
|----------------------|-----|------|-------|-------|-------|-------|-----|------|-----|------|-----|-------|
| <i>E. abyssinica</i> | 0   | 0    | 724   | 19741 | ND    | ND    | 0   | 0    | 174 | 4915 | ND  | ND    |
| <i>E. tirucalli</i>  | 0   | 0    | 0     | 12511 | 673   | 16435 | 0   | 0    | 79  | 3120 | ND  | ND    |
| <i>F. africana</i>   | 0   | 0    | 4883  | 15162 | ND    | ND    | 0   | 0    | 197 | 2091 | ND  | ND    |
| <i>Z. giletti</i>    | 0   | 0    | 27710 | 6643  | ND    | ND    | 0   | 0    | 120 | 654  | ND  | ND    |
| Negative control     | 745 | 2002 | 37611 | 3862  | 10597 | 18683 | 187 | 2957 | 212 | 5266 | 893 | 16017 |
| Potitive control     | 0   | 0    | 0     | 0     | 0     | 0     | 0   | 0    | 0   | 0    | 0   | 0     |

Key: GUs-Numerical growth units, Mk-*Mycobacteria kansasii*, Mtb-*M. tuberculosis*, Mf-*M. fortuitum*, Ms-*M. smegmatis*,

0-indicates complete inhibition, ND-Not done, Positive control- Isoniazid, Negative control-Dimethyl sulphoxide

\*Note: The higher the growth index, the less inhibitory the extract is to mycobacteria (Compared to negative control)

Table 2: Results of phytochemical screening of the plant extracts

| Botanical name of plant        | Tannins | Saponins | Flavonoids | Terpenoids | Cardiac glycosides | Alkaloids (Wagner's test) |
|--------------------------------|---------|----------|------------|------------|--------------------|---------------------------|
| <i>Carissa edulis</i>          | ++      | +        | +++        | +          | -                  | ++                        |
| <i>Croton macrostachyus</i>    | -       | +        | +          | -          | -                  | -                         |
| <i>Vernonia amygdalina</i>     | +       | +        | +++        | -          | -                  | +                         |
| <i>Momordica charantia</i>     | ++      | -        | +++        | +          | +                  | +                         |
| <i>Ocimum gratissimum</i>      | +++     | +        | +++        | ++         | ++                 | +++                       |
| <i>Lantana camara</i>          | +++     | +        | +++        | +          | ++                 | +                         |
| <i>Clerodendrum myricoides</i> | +++     | +        | +++        | +          | +                  | +++                       |
| <i>Toddalia asiatica</i>       | +       | -        | ++         | -          | -                  | +                         |
| <i>Lantana trifolia</i>        | +++     | ++       | ++         | -          | +                  | +                         |
| <i>Erythrina abyssinica</i>    | ++      | ++       | +++        | +          | -                  | +++                       |
| <i>Euphorbia tirucalli</i>     | +++     | -        | +++        | +++        | +                  | +                         |
| <i>Fuerstia africana</i>       | +       | -        | ++         | +++        | +                  | +                         |
| <i>Zanthoxylum giletti</i>     | -       | +        | +          | ++         | -                  | +                         |

**Key:** +++ = Present in high concentration, ++ = Moderately Present, + = Trace, = Absent

### **Candida albicans and other bacteria**

The extracts from the selected medicinal plants gave varying degrees of antimicrobial activities against the test microorganisms (Table 3). *Carissa edulis* had strong antimicrobial activity (zone of inhibition of 9.00 mm and both MIC and MBC of 37.5mg/mL) against *S. typhi* (Table 3 and 4). It could be possible that the alkaloids found in the roots played an important role in the medicinal value of the plant (11, 14, 18) (Table 2). *Toddalia asiatica* extract was active only against *P. aeruginosa* with a zone of inhibition of 10.66 mm and both MIC and MBC of 9.375 mg/mL, whereas *Lantana trifolia* extract which was not active against the mycobacteria strains, had strong antimicrobial activity against *S. aureus* and *P. aeruginosa* (zones of inhibition of 20mm and 10mm respectively, with MIC and MBC of 37.5 mg/mL in both cases). The extract was bactericidal against the two test cultures with both MICs and MBCs at 37.5mg/ml. This activity is possibly due to high content of essential oils such as monoterpenes and sesquiterpene (17).

*Momordica charantia* was active only against *S. aureus* (inhibition zone of 10.66 mm, MIC and MBC of 37.5mg/mL) and *P. aeruginosa* (inhibition zone of 9.33mm MIC and MBC of 37.5mg/mL) which indicated strong antimicrobial activity (Table 3 and 4). Activity against the two test cultures could be attributed to the presence of the flavonoids (Table 2). They could have also interfered with the integrity of lipopolysaccharides in *P. aeruginosa* (21). This is in agreement with the argument by Ushimaru *et al.*, (16), that medicinal plants are important for the study of their traditional uses through the verification of pharmacological effects and can be natural composite sources that act as new anti-infectious agents.

*Croton macrostachyus*, *Vernonia amygdalina* had only moderate to poor activity (zones of inhibition of 6.66mm-8.0mm) against the test cultures used for general antibacterial and antifungal screening, possibly due to the antagonistic nature of the phytochemicals (22) (Table 3 and 4). *Candida albicans* was resistant against all the test extracts. This could be due to phytochemicals antagonism (22).

Table 3: Zones of inhibition produced by plant extracts

| Botanical name of plant        | <i>S. typhi</i> | <i>S. aureus</i> | <i>E. coli</i> | <i>P. aeruginosa</i> | <i>K. pneumoniae</i> | <i>C. albicans</i> |
|--------------------------------|-----------------|------------------|----------------|----------------------|----------------------|--------------------|
| <i>Carissa edulis</i>          | 9.00            | 8.00             | 7.00           | 7.66                 | 8.00                 | 7.00               |
| <i>Croton macrostachyus</i>    | 8.00            | 7.66             | 7.00           | 7.33                 | 6.66                 | 8.00               |
| <i>Vernonia amygdalina</i>     | 7.33            | 8.66             | 6.66           | 8.33                 | 8.00                 | 7.66               |
| <i>Momordica charantia</i>     | 8.66            | 10.66            | 6.00           | 9.33                 | 8.00                 | 7.66               |
| <i>Ocimum gratissimum</i>      | 8.66            | 8.66             | 7.00           | 7.66                 | 6.66                 | 8.00               |
| <i>Lantana camara</i>          | 6.66            | 9.00             | 6.00           | 8.66                 | 7.33                 | 6.33               |
| <i>Clerodendrum myricoides</i> | 7.66            | 8.00             | 7.00           | 7.66                 | 6.66                 | 7.66               |
| <i>Toddalia asiatica</i>       | 6.33            | 8.66             | 7.00           | 10.66                | 6.00                 | 8.00               |
| <i>Lantana trifolia</i>        | 8.66            | 20.00            | 8.66           | 10.00                | 6.66                 | 6.66               |
| <i>Erythrina abyssinica</i>    | 8.00            | 8.66             | 7.33           | 10.33                | 8.00                 | 8.00               |
| <i>Euphorbia tirucalli</i>     | 6.33            | 6.33             | 7.66           | 6.33                 | 7.00                 | 7.00               |
| <i>Fuerstia africana</i>       | 7.33            | 8.00             | 7.66           | 6.66                 | 6.66                 | 7.00               |
| <i>Zanthoxylum giletii</i>     | 6.66            | 7.33             | 6.00           | 9.33                 | 7.00                 | 7.66               |
| Positive controls              | 16.00           | 21.33            | 20.22          | 17.33                | 17.66                | 13.00              |
| Negative controls              | 6.00            | 6.00             | 6.00           | 6.00                 | 6.00                 | 6.00               |

#### Controls

The Negative controls were made from Methanol discs (evaporated).

The positive control for *Candida albicans* was Fluconazole

The positive control for *Salmonella typhi* and *Klebsiella Pneumoniae* was Zeftazidime and Ciprofloxacin respectively.

The positive control for *Staphylococcus aureus*, *Escherichia coli* and *Pseudomonas aeruginosa* was Amoxicillin

Table 4: Minimum Inhibitory Concentrations (MICs) (mg/ml) and Minimum Bactericidal/fungicidal concentrations (MBCs/MFCs) (mg/ml) produced by the medicinal plants against bacterial and fungal test cultures

| Botanical name              | <i>S. typhi</i>              |       | <i>S. aureus</i> |       | <i>E. coli</i> |       | <i>P. aeruginosa</i> |       |
|-----------------------------|------------------------------|-------|------------------|-------|----------------|-------|----------------------|-------|
|                             | MIC                          | MBC   | MIC              | MBC   | MIC            | MBC   | MIC                  | MBC   |
| <i>Carissa edulis</i>       | 37.5                         | 37.5  | ND               | ND    | ND             | ND    | ND                   | ND    |
| <i>Lantana camara</i>       | ND                           | ND    | 37.5             | 37.5  | ND             | ND    | ND                   | ND    |
| <i>Toddalia asiatica</i>    | ND                           | ND    | ND               | ND    | ND             | ND    | 9.375                | 9.375 |
| <i>Lantana trifolia</i>     | ND                           | ND    | 37.5             | 37.5  | ND             | ND    | 37.5                 | 37.5  |
| <i>Momordica charantia</i>  | ND                           | ND    | 37.5             | 37.5  | ND             | ND    | 37.5                 | 37.5  |
| <i>Erythrina abyssinica</i> | ND                           | ND    | ND               | ND    | ND             | ND    | 37.5                 | 37.5  |
| Positive controls           | 4.687                        | 4.687 | 4.687            | 4.687 | 4.687          | 4.687 | 4.687                | 4.687 |
| Negative controls           | Growth observed in all tubes |       |                  |       |                |       |                      |       |

Key: ND- Not done, Mean MIC values done in triplicate,

The results from present study validate and document, in a systematic way, that some of the plant species studied possess substantial antimicrobial properties. This explains the use of some of them in folk medicine against various diseases. Further study is underway for purification, separation, isolation and characterization of the active principles from the active extracts.

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Appendix 1: Selected medicinal plants used by the communities around the Lake Victoria region to treat mycobacterial, selected bacterial and fungal diseases

| <b>Botanical Name</b>                                     | <b>Family name</b> | <b>Voucher specimen no.</b> | <b>Local name(s)</b>                      | <b>Where collected from</b> | <b>Part(s) used</b> | <b>Diseases treated</b>                           |
|---|--------------------|-----------------------------|---|-----------------------------|---------------------|---|
| <i>Carissa edulis</i> Vahl                                | Apocynaceae        | TB/MR/028/07                | Omonyangateti (Kisii)                     | Kisii South                 | Roots               | Polio, TB, asthma, Gonorrhoea, Malaria            |
| <i>Croton macrostachyus</i> Hochst. ex Ferret et Galinier | Euphorbiaceae      | TB/MR/008/07                | Odhwidho (Luo)<br>Omosocho (Kisii)        | Siaya<br>(Alego)            | Stem<br>bark        | Stomach ache, TB, asthma, coughs, rheumatism      |
| <i>Vernonia amygdalina</i> Del.                           | Asteraceae         | TB/MR/017/07                | Okelo – okelo (Luo)<br>Omosabakwa (Kisii) | Kisii south                 | Root<br>barks       | gastrointestinal problems, TB, Asthma             |
| <i>Momordica charantia</i> L.                             | Curcubitaceae      | TB/MR/058/07                | Echokilayiti (Teso)                       | Teso                        | Whole plant         | Asthma, TB, pneumonia                             |
| <i>Ocimum gratissimum</i> L.                              | Labiatae           | TB/MR/016/07                | Obweny (Luo)                              | Siaya                       | Leaves              | Ear problems, chest pains                         |
| <i>Lantana camara</i> L.                                  | Verbaenaceae       | TB/MR/019/07                | Nyamndhi<br>(Nyabende/ Atek)<br>(Luo)     | Bondo<br>(Sakwa)            | Leaves              | Ulcers, TB, Pneumonia, chest pains, Malaria       |
| <i>Clerodendrum myricoides</i> (Hochst)Vatke              | Verbenaceae        | TB/MR/057/07                | Okwerogweno (Luo)<br>Omonyasese (Kisii),  | Siaya<br>(Alego)            | Roots               | Gonorrhoea , Mumps, TB, Chest problems, hepatitis |

|  |               |              |  |                  |               |   |
|--|---------------|--------------|--|------------------|---------------|---|
| <i>Toddalia asiatica</i> (L.) Lam.             | Rutaceae      | TB/MR/022/07 | Nyalwet kwach (Luo)<br>Ekenagu ekiegarori<br>(Kisii) | Bondo<br>(Alego) | Roots         | Stomach ache,<br>Measles, TB,               |
| <i>Lantana trifolia</i> L.                     | Verbenaceae   | TB/MR/023/07 | Nyabendwiny (Luo)<br>Obori bwenyoni<br>(Kisii)       | Bondo<br>(Sakwa) | Leaves        | Heart problems,<br>Gonorrhea, TB,<br>coughs |
| <i>Erythrina abyssinica</i><br>Lam ex DC       | Fabaceae      | TB/MR/007/07 | Omotembe (Kisii)                                     | Kisii<br>Central | Root<br>barks | TB, Malaria, Stomach<br>ache                |
| <i>Euphorbia tirucalli</i> L.                  | Euphorbiaceae | TB/MR/021/07 | Ekerachwoki (Kisii)                                  | Kisii<br>Central | Stem          | TB, asthma                                  |
| <i>Fuerstia africana</i><br>T.C.E.Fr.          | Lamiaceae     | TB/MR/002/07 | Mienya (Luo)<br>Ekebunga baiseke<br>(Kisii)          | Bondo<br>(Sakwa) | Leaves        | Malaria, Asthma,<br>chest problems          |
| <i>Zanthoxylum giletti</i><br>De Wild.) Waterm | Rutaceae      | TB/MR/010/07 | Egekoma<br><br>(Kisii)                               | Kisii<br>Central | Stem<br>bark  | TB, Asthma                                  |

## References

1. JG Gotep, GOA Agada, DS Gbise, S Chollom. Antibacterial activity of ethanolic extract of *Acalypha wilkesiana* leaves growing in Jos, Plateau State, Nigeria. *Mal. J. Microbiol.* 2010, 6(2), 69-74.
2. J Cohen MS Dibner, A Wilson. Development of and Access to Products for Neglected Diseases. PLoS ONE 2010, 5(5), e10610 doi: 10.1371.
3. AM Marchi, ID Juttel, EM Kawacubo, EM Dalmarco, SL Blatt, CMM Cordova. Evaluation of methods for detection and identification of mycobacterium species in patients suspected of having pulmonary tuberculosis. *Braz. J. Microbiol.* 2008, 39, 613-618.
4. JGB Sanchez, VV Kouznetsov. Antimycobacterial susceptibility testing methods for natural products research. *Braz. J. Microbiol.* 2010, 41, 270-277.
5. R Khan, B Islam, M Akram, S Shakil, A Ahmad, SM Ali, M Siddiqui, AU Khan. Antimicrobial Activity of Five Herbal Extracts Against Multi Drug Resistant (MDR) Strains of Bacteria and Fungus of Clinical Origin. *Molecules* 2009, 14, 586-597.
6. SK Panda, S Brahma, SK Dutta. Selective antifungal action of crude extracts of *Cassia fistula* L.: A preliminary study on *Candida* and *Aspergillus* species. *Mal. J. Microbiol.* 2010, 6(1), 62-68.
7. NL Alencar, TADS Araujo, ELCD Amorim, UPD Albuquerque. The inclusion and selection of medicinal plants in traditional pharmacopoeias-evidence in support of the diversification hypothesis. *Econ. bot.* 2010, 64 (1), 68-79.
8. S Johann, MG Pizzolatti, CL Donnici, MA Resende. Antifungal properties of plants used in Brazilian traditional medicine against clinically relevant fungal pathogens. *Braz. J. Microbiol.* 2007, 38, 632-637.

9. MA Resende, S Johann, MG Pizzolatti, CL Donnici. Antifungal properties of plants used in Brazilian traditional medicine against clinically relevant fungal pathogens. *Braz. J. Microbiol.* 2007, 38, 632-637.
10. OO Aiyelaagbe, PM Osamudiamen. Phytochemical screening for active compounds in *Mangifera indica* leaves from Ibadan, Oyo State. *Plant Sci. Res.* 2009, 2(1), 11-13.
11. HO Edeoga, DE Okwu, BO Mbaebie. Phytochemical constituents of some Nigerian medicinal plants. *Afr. J. Biotechnol.* 2005, 4 (7), 685-688.
12. L Srisuwanvilai, P Monkongdee, LJ Podewils, K Ngamlert, V Pobkeeree. Performance of the BACTEC MGIT 960 compared with solid media for detection of Mycobacterium in Bangkok, Thailand. *Diagnostic Microbiology and Infectious Disease* 2008, 61, 402-407.
13. Becton, Dickinson Company. BBL MGIT Mycobacteria growth indicator Manual. Maryland, USA. 2007, 1-23.
14. J Parekh, SV Chanda. *In vitro* Antimicrobial Activity and Phytochemical Analysis of Some Indian Medicinal Plants. *Turk. J Biol.* 2007, 31, 53-58.
15. P Rani, N Khullar. Antimicrobial Evaluation of some Medicinal Plants for their Anti-enteric Potential against Multi-drug Resistant *Salmonella typhi*. *Phytother. Res.* 2004, 18, 670-673.
16. PL Ushimaru, MTN Silva, LC Stasi, L Barbosa, AF Junior. Antibacterial activity of medicinal plant extracts. *Braz. J. Microbiol.* 2007, 38, 717-719.
17. OC Nwinyi, NS Chinedu, OO Ajani, CO Ikpo, KO Ogunniran. Antibacterial effects of extracts of *Ocimum gratissimum* and *piper guineense* on *Escherichia coli* and *Staphylococcus aureus*. *Afr. J. Food Sci.* 2009, 3(3), 77-81.

18. AK Brown, A Papaemmanouil, V Bhowruth, A Bhatt, LG Dover, GS Besra. Flavonoid inhibitors as novel antimycobacterial agents targeting Rv0636, a putative dehydratase enzyme involved in *Mycobacterium tuberculosis* fatty acid synthase II. *Microbiology* 2007, *153*, 3314-3322.
19. E Yuan, B Liu, Z Ning, C Chen. Preparative separation of flavonoids in *Adinandra nitida* leaves by high-speed counter-current chromatography and their effects on human epidermal carcinoma cancer cells. *Food Chemistry* 2009, *115*, 1158-1163.
20. Y Cheng, J Pieters. Novel proteasome inhibitors as potential drugs to combat tuberculosis. *J. Mol. Cell Bio.* 2010, doi: 10.1093.
21. KE Mdluli, PR Witte, T Kline, AW Barb, AL Erwin. Molecular Validation of LpxC as an Antibacterial Drug Target in *Pseudomonas aeruginosa*. *Antimicrob. Agents and Chemother.* 2006, *50*(6), 2178-2184.
22. EK Ruttoh. *Antimicrobial efficacy of selected plants use by traditional medical practioners in Kerio, Kenya.* Nairobi, Kenya. M.Sc. Dissertation. Kenyatta University, 2009, 78-87.

**\* For Corresponding**

Prof. Paul O. Okemo,

Email: [okemo1952@yahoo.com](mailto:okemo1952@yahoo.com)

Cell: +254-722942072

Fax: +254-020-810759