



Preliminary Medicinal Value Evaluation of Some Plants Used by the Ogiek in Management of Microbial Related Infections

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Authors' contributions

This work was carried out in collaboration between all authors. The work was designed by the first author, OA. He identified the voucher specimens and they were authenticated by National Museums of Kenya and PM, the extractions were done by authors OA and AKM. The Microbial studies were carried out by authors OA and POO finally the drafting was done by author OA and the editing by everybody. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/EJMP/2015/16295

Editor(s):

(1) Marcello Iriti, Professor of Plant Biology and Pathology, Department of Agricultural and Environmental Sciences, Milan State University, Italy.

Reviewers:

(1) Said Laarabi, Biology, University Mohammed V, Morocco.

(2) Anonymous, University Hassan II, Morocco.

(3) Anonymous, The University of Hong Kong, China.

Complete Peer review History: <http://sciencedomain.org/review-history/9964>

Original Research Article

Received 22nd January 2015

Accepted 26th April 2015

Published 29th June 2015

ABSTRACT

Aim: The survey was targeted at documenting the indigenous plants that are used in the management of microbial related medical conditions within the Ogiek communities of Kenya.

Study Design: Purposive sampling approach was used to interview the traditional herbalist with the help of a questionnaire.

Place and Duration of the Study: Some 49 plants species used by the Ogiek, who are indigenous forest and forest product dwellers and users, in the management of microbial related conditions in human maladies were collected from the Mau Forest complex and prepared, for extractions and screened for antimicrobial activities at Kenyatta University and National Public Health Laboratories, Nairobi respectively. The plants were screened to ascertain their activities against selected human

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microbial infections.

Results: Crude methanol extracts from 16 species showed activities against various pathogenic organisms. There was also marked activities from two of the oils extracted from three species of the 16 plants.

Conclusion and Recommendations: Some plants have activities against selected microorganisms and this validates their continued use as medicine by the Ogiek communities and their neighbours as medicine. It has been suggested that further studies be done on the bioactive plant species to establish their bactericidal and fungicidal abilities; and their safety to humans to justify their continued use as medicine.

Keywords: Ogiek; medicinal plants; antimicrobial activities; Kenya.

1. INTRODUCTION

The Ogiek of the Rift Valley in Kenya are some of the oldest indigenous people of Sub-Saharan Africa and are a rich source of ethno-botanical information (Towett, 2002). They have led a secretive life in the Tropical Montane Forests of Central Rift Valley as hunters and wild plant product gatherers. The tasks of documenting their vast and conservative ethnology are urgent because this indigenous knowledge is not properly documented. Furthermore, their natural habitats have been encroached by surrounding communities. Unfortunately the ecosystems of forests are fragile and are getting disseminated because of population pressure. Consequently, there is an accelerated reduction in the biodiversity in such areas. Ogiek are some of the oldest indigenous inhabitants in sub-Saharan Africa that feel threatened by these new developments

The purpose of carrying out the antimicrobial screening was meant to uncover and verify the efficacy of the plants which are currently in use and to uncover new antimicrobial agents from higher plants, thus exposing their potentials. It was also aimed at demystifying the secrecy in which traditional African medicine has been shrouded for centuries. In the past, there has been no scientific verification to validate the efficacy of the traditional Phytomedicine in the Ogiek community. This is what the current screening and conventional laboratory studies is set to carry out to an extent that only promising ones may be recommended for continued therapeutic uses in the traditional setups. The community gives virtually every account of any abnormal conditions both in human and their livestock that may manifest itself as an ailment.

2. MATERIALS AND METHODS

49 medicinal plants were collected from the Ogiek traditional healers who live in the Mau

Forest Complex in the Rift Valley. The area has Afromontane Flora covering the parts in which these people live. Using purposive sampling method, 20 traditional healers were interviewed, and the plant species identified and voucher specimens prepared and deposited at the National Museums of Kenya – East African Herbarium (EAH) for verification. The plant species that were used because of their bioactivities are: *Olea europaea* L. ssp. *Africana* (Mill.) P. Green (Yemdit), *Rhamnus pirinoides* (Kosisito), *Olea capensis* L. (*O. hochstetteri*) (Maseita), *Faurea saligna* Harv. (Msomboriet), *Ekabergia capensis* Sparrm. (Ororuet), *Bersama abyssinica* Fres. (Sagaweita), *Scutia myrtina* (Burm.f.) Kurtz (Sumbeyet), *Clusia robusta* Pax (Kibaryaat), *Toddalia asiatica* (L.) Lam. (Chepindoruet), *Impatiens tinctoria* A. Rich. (Pumbutiet), *Lippia Javanica* (Burm.f.) Spreng (Mwokyot), *Satureia biflora* (D. Don.) Benth. (Chepsagitiet), *Embelia schimperi* Vatke (Sachuonet), *Olinia rochetiana* A. Juss. (Kabideleliet) and *Rubus keniensis* Standl. (Tagameit). Plant parts used as medicines were collected, methods of administration recorded, and any other pharmacological profiles. The samples were dried at room temperature of 21°C. The powder, of each sample, was hermetically sealed in polythene bags, and stored till the time of use if not extracted immediately. The samples were extracted with analytical grade methanol for 24 hours.

2.1 Methanol Extraction

The plant powders from the 49 plants were individually extracted with methanol while fresh leaves were steam distilled. Some powdered material (50 g) were weighed into conical flasks (250 ml), covered with aluminum foil and then filled with about 200 ml of methanol. This was allowed to stand for 24 hrs.

The solution was then decanted and filtered through Whatman filter paper No 1. The filtrate

was transferred into aluminum-foiled beakers and allowed to evaporate at room temperature to dryness. Various forms of residues were obtained as crude extracts. The resultant materials were put into individual vials labelled and stored in the refrigerator at 4°C for future use.

2.2 Reconstitution of the Test Samples

Each of 0.1 g individually dried material was dissolved in about four drops of dimethyl sulphoxide (DMSO), so as to make it absorbable by the test organisms, and topped up to 1 ml. of water. Discs made from what man filter paper no. 1 and measuring 6 mm in diameter, previously sterilized, and were directly dipped into the solution, removed, desiccated in Silica gel and later placed onto the plate. With the help a micro-syringe, required amounts of Essential oils were also individually loaded onto the sterilized paper discs at the rate of 0.1 ml in the case of methanol extracts [1].

2.3 Screening for Antimicrobial Activity

Antimicrobial efficacies were tested using the standard filter paper disc diffusion method [1]. The Muller-Hinton and Potato Dextrose Agar (PDA) were used in the culture of bacteria and fungi, respectively. The bacterial and fungal cultures were incubated at 37°C for 24 to 48 hrs, respectively, while fungi were incubated at 25°C for 5 days. After the incubation period the zones of inhibition were measured and recorded in mm as described by [1]. Negative control plates had discs with sterile methanol. The plants which were found to possess bioactivities were further subjected to screening to ascertain their zones of inhibition at various concentrations. Antimicrobial sensitivity and resistance were confirmed by use of standard discs containing ampicillin (10 µg), chloramphenicol (30 µg), erythromycin (15 µg), gentamycin (10 µg), ciprofloxacin (10 µg), tetracycline (30 µg), amikacin (30 µg) and an additional oxacilin (1 µg) for *S. aureus* (oxoid, London). *Candida albicans*, *Candida brusei* and *Cryptococcus neoformans*, *richophyton mentagrophytes* and *Microsporium gypseum* were incubated at 25°C for a period of five days to ascertain the activities of the extracts their standards for fungi were discs containing fluconazole. Broths without any treatments and inoculations were taken as positive controls. All the pathogens used were based on the common occurrence in medical conditions.

3. RESULTS AND DISCUSSIONS

3.1 Methanol Extracts

The results presented in the subsequent tables indicate the bioactivities of various plant species. Table 1 shows a general view of plants that act on bacteria alone. There was also a category of plants which possess activities on both bacteria and fungi. For both categories of pathogens, a total of 16 plants were found to be active. Of the 49 plant species only 16 showed activities against both bacteria and fungi. Using a ruler, the zones of inhibition were measured across each petri dish. The + sign was used to indicate any sensitivity to the drugs. A single + indicated 8 mm inhibition in diameter from the edge of the disc., >9 but 15 mm ++ indicated moderate activity and >16 mm +++ high activity, while – indicating no reactions. The same procedure above in bacteria was followed for the investigations into the pathogenic fungi which are found in humans. The extracts that were found to be biologically active were further subjected to screening at different concentrations and stains of the test organisms. The results of the antifungal tests have been presented in the Table 1 with the preliminary results for the bacteria.

The *Salmonella* spp. also exhibited resistance to the methanol extracts of *B. abysinica*, and to most other test drugs. The reaction also included all the strains of the sub-cultures of the pathogen that are listed herein: *Salmonella* (*S. mim*), *S. typhi*, *S. Group D* (Clinical isolates) *S. typhi* types and *R. colidale* amp, Similar results were obtained from methanol extracts from *Bersama* stem bark extracts with the strains of: *E. coli* (0125/B15), *E. coli* (086), *E. coli* (0125 K70), *E. coli* (0127/8), *E. coli* (D126/B16), and *E. coli* (087/B7). However, several extracts from various species demonstrated efficacies against the selected microbial pathogens as can be seen in the Tables.

Table 1 the plants of methanol extracts that possessed antimicrobial properties were further subjected to the same organisms but at different serial concentrations and exhibited increase in the areas of inhibitions. Zones of inhibition varied from one organism to the other (Figs. 1 to 4). The subsequent Tables 2 to 14 contains individual extracts efficacies with quantum concentrations against different organisms including their sub cultures. Some of the sub cultures are universally known resistant to current broad spectrum

antibiotics. The plants crude methanol extracts showed good antimicrobial activities on them.

There was resistance on fungi against the majority of the plant extracts with a few showing activities. This could be attributed to the fact that the drugs are able to penetrate the organisms' cell wall. The inability may be, due to the fact that fungi being plants, though from the lower classes may have similar compounds as the extracts. *S. biflora* was found to be effective on *K. pneumoniae*, *S. aureus*, *P. aeruginosa* and *E. coli*. *F. saligna* was effective on *E. coli*, *S. typhi* and *E. faecalis* while *B. abyssinica* was effective on *K. pneumoniae*, *S. aureus*, *E. coli* and *C.*

albicans (Table 9) with St Dev= 0,456 which was significantly different. There were variable activities of individual methanol extracts from one species to the other.

Inhibition of *E. coli* strains with *O. rochetiana*, though concentrations, showed the same trend in increase and finally stabilizing at 20 mm for *O. rochetiana* and 10mm for *Olea* spp. as seen in Fig. 3.

Inhibition of *Staphylococcus* spp. showed varied but minimally to changes in concentrations, Fig. 4.

Table 1. Screening for activity

Name of plants	S.a.	P.a.	E.f.	K.p.	E.c.	S.t.	C.a.	C.b.	C.n.	T.m.	M.g.
<i>R. prinoides</i>	++	-	-	+	-	-	-	-	-	-	-
<i>O. e. ssp. africana</i>	++	-	++	+	-	-	-	-	-	-	-
<i>O. hochstetteri</i>	+++	-	-	+	-	-	-	-	-	-	-
<i>F. saligna</i>	+++	++	-	++	-	-	+++	+++	+++	++	+++
<i>E. capensis</i>	+++	+++	++	++	-	-	-	-	-	-	-
<i>B. abyssinica</i>	+++	-	+	+++	-	-	++	+++	++	+++	++
<i>S. myrtina</i>	+++	++	-	+++	-	-	-	-	-	-	-
<i>G.buchananni</i>	++	-	-	-	-	-	-	-	-	-	-
<i>C. robusta</i>	+++	++	+	+++	-	-	-	-	-	-	-
<i>T. asiatica</i>	+++	-	-	-	-	-	+++	+++	+++	++	++
<i>I. tinctoria</i>	++	-	-	-	-	-	++	++	+++	+	++
<i>L. javanica</i>	++	-	+	-	-	-	+	+	+	-	-
<i>S. biflora</i>	+++	+	+	-	+++	-	+++	+++	-	++	+
<i>E. schimperi</i>	+++	+++	+++	++	++	+	-	-	-	-	-
<i>O. rochetiana</i>	+++	+++	+++	+++	-	+++	-	-	-	-	-
<i>R. keniensis</i>	++	+++	+	++	-	+	-	-	-	-	-

Legend: *Staphylococcus aureus* (S.a.) *Pseudomonas aeruginosa*(P.a.) *Klebsiella pneumoniae* (K.p.) *Escherichia coli* (E.c.) *Salmonella typhi* (S.t.) *Candida albicans* (C.a.) *Candida brusei* C.b. *Cryptococcus neoformas* (C.n) *Trichophyton mentagrophyte*(T.m.) *Microsporium gypseum* (M.g.); +++ High activity ++ moderate activity + slight activity - no activity

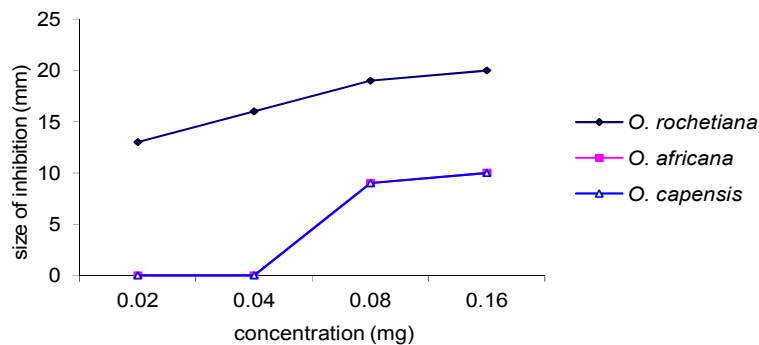


Fig. 1. various inhibitions in mm of methanol extracts against *E. coli*

Klebsiella spp. did not show susceptibility despite the increase in concentrations of the methanol extracts except in *O. africana* stem bark methanol extracts which had a sharp increase between (Fig. 4).

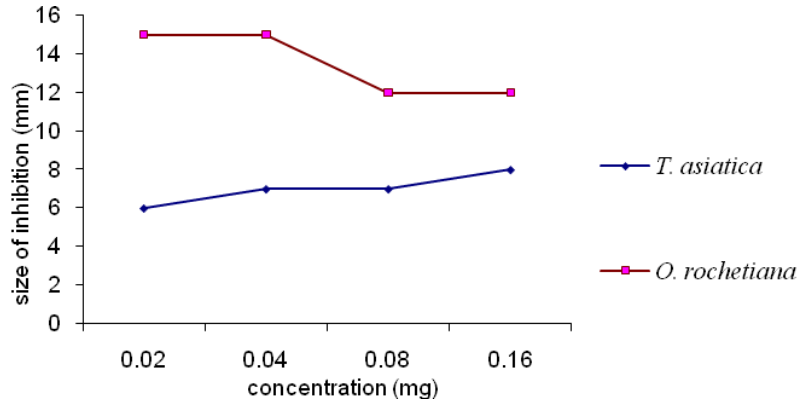


Fig. 2. Stem bark methanol extracts of *T. asiatica* and *O. rochetiana*

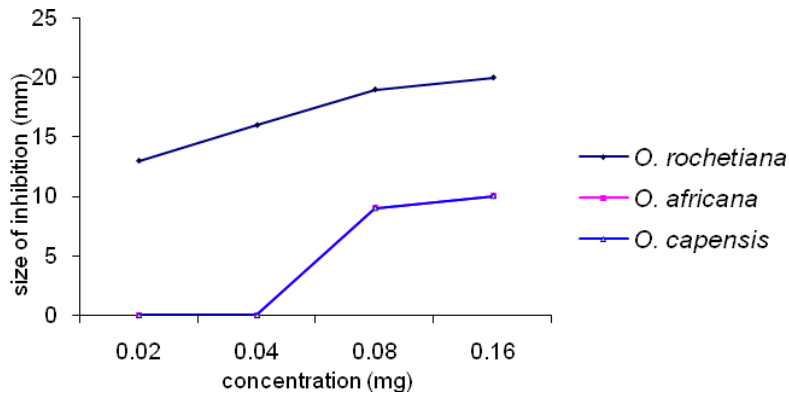


Fig. 3. Stem bark methanol extracts of *O. rochetiana*, *O. africana* and *O. hochstetteri*

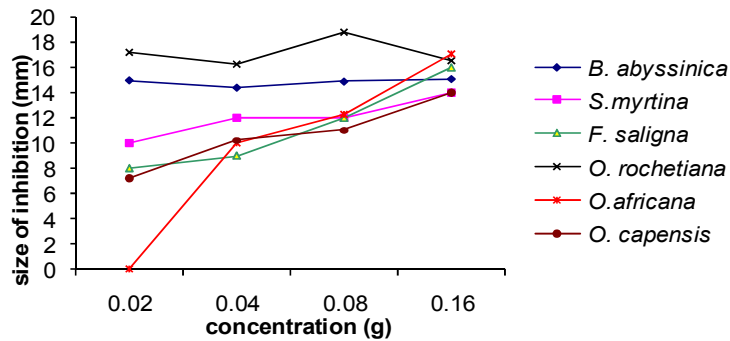


Fig. 4. Stem bark extracts of various drugs inhibition of *Klebsiella* spp

The subsequent Tables show various activities of plants various extracts on human microbial pathogens.

Table 2: The following strains of *Pseudomonas* used and all had similar reactions *P. aeruginosa*, *P. aeruginosa* ATCC27853 and *P. aeruginosa* (clinical isolates).

Table 2. Zones of inhibition (mm) by methanol extracts of *B. abysinica* against strains of *S. aureus* and *P. aeruginosa*

Pathogens	Conc. mg/ml			
	20	40	80	160
<i>S. aureus</i> (Pigmented with staphylokinase)	15	15	15	15
<i>S. aureus</i> (ATCC 20591)	15	14	15	15
<i>S. aureus</i> (ORSA) and MRSA	15	14	15	15
Strains of <i>P. aeruginosa</i>	10	10	10	10
Control	6	6	6	6

There was moderate inhibition of various strains of *S. aureus* by the extract (Table 3).

There was minimal to moderate susceptibility of *K. pneumoniae* and its strains to the *F. saligna* (Table 4).

There was no significant inhibition of the extract on *E. coli* while *S. aureus* showed high antimicrobial activity of the methanol extract against (Table 5).

There was moderate antimicrobial activity of the methanol extract against *Pseudomonas* spp, little activity against *S. typhi*, and high activity against strains of *E. faecalis* (Table 6).

There were variable activities of the methanol extract of *R. keniensis* against various strains of *P. aeruginosa* *E. faecalis*, *K. pneumoniae* and *S. aureus* (Table 7).

There were variable activities of the methanol extract, root bark, of *C. abysinica* against various strains of *E. faecalis*, *S. aureus*, *S. typhi*, *P. aeruginosa* and *K. pneumoniae* (Table 8).

At the similar concentrations similar zones of inhibition were realized from three different organisms listed in Table 9. The positive control had no growth.

At the similar concentrations similar zones of inhibition were realized from three different organisms listed in Table 10. The positive control had no growth.

Only *E. faecalis* (ATCC 29212) and *E. faecalis* (clinical isolates) were used in the study (Table 11).

Essential oils from *S. biflora* and *L. javanica* had similar activities (Table 12).

Activities of the organism were achieved in the last two plants extracts. The positive control had no growth (Table 13).

Several medicinal plant species yielded methanol extracts which were screened against six bacterial pathogens and three fungal species. There were a total of 49 plant species, out of which 16 species. 32.6% of the plant methanol extracts inhibited *S. typhi*, 12.1% of all the methanol extracts inhibited growth of *S. aureus*, 6.8% and *P. aeruginosa*. Extracts from *S. biflora*, *F. saligna* and *B. abysinica* were effective against most of the bacteria and *C. albicans*. *C. neoformas* and *T. mentagrophyte*. *S. biflora* were found to lightly inhibit, *S. aureus*, and *P. aeruginosa* and high inhibition of *E. coli*. *F. saligna* highly inhibited *S. aureus*, moderately *P. aeruginosa* and *K. pneumoniae*. The extract from the plant were also inhibitory on the test fungi while *B. abysinica* were highly inhibiting the; *K. pneumoniae*, *S. aureus*, *E. faecalis* and all the test fungi.

Table 3. Zones of inhibition (mm) by methanol extracts of *S. myrtina* stem bark against strains of *Klebsiella pneumonia*

Pathogens/ strains	Conc.20	in40	Mg/mg/80	ml160
<i>K. pneumoniae</i> (Clinical isolate)	11	11	14	15
<i>K. pneumoniae</i> (Belgium Strain)	11	12	16	16
<i>K. pneumoniae</i> (WHO)	11	11	14	15
<i>S. aureus</i> (Clinical isolate)	8	12	12	14
<i>S. aureus</i> (ATCC 20591)	8.0	12	12	14
<i>S. aureus</i> (haemolytic strain)	12	12	12	14
<i>S. aureus</i> (Pigmented strain)	12	12	12	14
Control	6	6	6	6

B. abysinica stem bark had extracts showed significant effect of inhibition against *K. pneumoniae* at various levels of conc. $P= 0.045$ suggesting that these results confirm the Ethnobotanical surveys whereby the plant is used for various ailments in human like fever and rheumatism and livestock east coast fever. Although the extract was active against *S. aureus* there was not much difference in response to increase in concentrations in the case of *Klebsiella spp.*

Table 4. Zones of inhibition (mm) by of *R. pirinoides* methanol extract against *S. aureus* and its strains

Pathogens	Conc.in mg/ml			
	20	40	80	160
<i>S. aureus</i> (Clinical isolate)	8	12	12	14
<i>S. aureus</i> (ATCC 20591)	8	12	12	14
<i>S. aureus</i> (haemolytic strain)	12	12	12	14
<i>K. pneumoniae</i> (WHO std)	8	12	12	14
<i>K. pneumoniae</i> (MDRS)	8	12	12	14
<i>K. pneumoniae</i> (Clinical isolates)	8	12	12	14
Control	6	6	6	6

Table 5. Zones of inhibition (mm) by *T. asiatica* root bark methanol extracts against strains of *S. aureus*

Pathogens	Conc. in mg/ml			
	20	40	80	160
<i>S. aureus</i> (Clinical isolate)	-6	7	12	16
<i>S. aureus</i> (β Haemolytic)	-6	7	14	16
<i>S. aureus</i> (Pigmented)	-6	7	12	16
<i>S. aureus</i> (Pigmented with staphylokinase)	6-	7	12	16
<i>S. aureus</i> (ATCC 20591)	-6	7	13	17
<i>S. aureus</i> (ORSA)	-6	7	14	16
<i>S. aureus</i> (MRSA)	-6	7	12	16
<i>E. coli</i> (std)	6	7	7	8
<i>E. coli</i> (std 25922)	6	7	7	8
<i>E. coli</i> (35218)	6	7	7	8
<i>E. coli</i> (0125/B15)	6	7	7	8
<i>E. coli</i> (087/B7)	6	7	7	8
<i>E. coli</i> (0126/B16)	6	7	7	8
<i>E. coli</i> (125/K70)	6	7	7	8
<i>E. coli</i> (0127/8) control	6	7	7	8

Table 6. Zones of inhibition (mm) by methanol extracts *O. rochetiana* against strains of: *E. faecalis* spp *P. aeruginosa* and *S. typhi*

Pathogens	Conc in mg/ml			
	20	40	80	160
<i>E. faecalis</i> (clinical isolates)	13	16	19	20
<i>E. faecalis</i> ATCC 29212	13	16	19	20
<i>P. aeruginosa</i> (Clinical isolates)	10	10	12	12
<i>P. aeruginosa</i> ATCC 27853	10	10	12	12
<i>S. paratyphi</i>	6	10	10	10
<i>S. typhi</i> (type R. colidale amp)	6	10	10	12
<i>S. enteridis</i>	6	10	11	11
<i>Salmonella</i> (clinical isolate)	6	10	10	10
<i>S. typhi</i> N2202	6	10	10	10
Control	6	6	6	6

Table 7. Zones of Inhibition (mm) by *R. keniensis* root bark methanol extracts *P. aeruginosa*, *K. pneumoniae* and *E. faecalis*

Pathogens	Conc in mg/ml			
	20	40	80	160
<i>P. aeruginosa</i> (Clinical isolates)	10	16	20	22
<i>P. aeruginosa</i> ATCC 27853	10	15	20	22
<i>E. faecalis</i>	6	10	7	10
<i>K. pneumoniae</i>	7	10	12	14
<i>S. aureus</i>	7	12	14	15
Control	6	6	6	6

The $P= 0.040$ in the case *S. aureus*, *B. abysinica* is not mentioned in other literature as a medicinal plant of significance. However, in East Africa several communities use it to treat colds, aphrodisiac, purgative emetic and anti diarrhea [2]. The records on its efficacy and bioactivities are limited in literature. The information on efficacy on growth inhibition at 50% at concentration 0.08 gm/ml, against *K. pneumoniae* (Fig. 4) and this could be the reason for its use as a remedy for colds and chest related complaints by the community in the study. The experience of inhibition was the same in case of *S. aureus* and its strains.

S. myrtina and *R. prinoidea*, Rhamnaceae both are used as medicines. The latter is prepared by incorporating the root bark in cookery, and the former stem bark as tea tonic and treatment of

chest ailments. There was a significant effects, 12 mm and 15mm, inhibitions, of *S. myrtina* extracts at the concentration used in the studies, at 100% P= 0.043 in inhibiting growth of *P. aeruginosa* at 160 mg/ml (Table 3).

Table 8. Zones of inhibition (in mm) by methanol extracts of root bark of *C. abyssinica* against *E. faecalis*

Pathogens	Conc in mg/ml			
	20	40	80	160
<i>E. faecalis</i> (clinical isolates)	6	7	10	13
<i>E. faecalis</i> ATCC 29212				
<i>S. aureus</i> (Clinical isolate)	7	7	11	16
<i>S. aureus</i> (β Haemolytic)	7	7	11	15
<i>S. aureus</i> (Pigmented)	7	7	11	16
<i>S. aureus</i> (Pigmented with staphylokinase)	7	7	11	16
<i>S. aureus</i> (ATCC 20591)	7	7	11	16
<i>S. aureus</i> (ORSA)	7	7	11	16
<i>S. aureus</i> (MRSA)	7	7	11	16
<i>S. paratyphi</i>	6	7	7	10
<i>S. typhi</i> (type R. colidale amp)	6	7	9	10
<i>S. enteridis</i>	6	7	7	10
<i>Salmonella</i> (Clinical isolate)	6	7	8	11
<i>S. typhi</i> N2202	6	7	7	11
<i>P. aeruginosa</i> (Clinical isolates)	7	9	10	11
<i>P. aeruginosa</i> ATCC 27853	7	9	10	11
<i>K. oxytoca</i>	6	11	12	15
<i>K. pneumoniae</i> (clinical isolate)	6	12	12	16
<i>K. pneumoniae</i> (Belgium stain)	6	11	13	15
<i>K. pneumoniae</i> (WHO std)	6	12	12	16
<i>K. pneumoniae</i> (MDRS)	6	11	12	14
Control	6	6	6	6

Table 9. Zones of inhibition (mm) by *G. burchananni* methanol leaf extract against *S. aureus* and *E. faecalis*

Organism	Conc. (100 mg/ml)
<i>S. aureus</i> (ORSA)	10
<i>S. aureus</i>	10
<i>C. albicans</i>	10
Control	6

Table 10. Zones of inhibition (mm) by *I. tinctoria* 100 mg/ml root methanol extract

Organism	Conc. (100 mg/ml)
<i>S. aureus</i>	25
<i>S. aureus</i> (ORSA)	10
<i>B. subtilis</i>	10
Control	6

Table 11. Zones of inhibition (mm) by drug conc.100 mg/ml *I. tinctoria* root methanol extract against *E. faecalis*

Conc. in mg/ml	Conc. (100 mg/ml)
25	12
50	14
100	18
200	20
Control	6

Table 12. Zones of inhibition (mm) by *S. biflora* essential oils activities (100 mg/ml)

Organism	Zones of inhibition (mm)	Control (mm)
<i>S. aureus</i> (ORSA)	19	6
<i>S. aureus</i>	10	6
<i>K. pneumoniae</i>	10	6
<i>K. pneumoniae</i> (MDRS)	11	6
<i>E. coli</i>	17	6

Table 13. Zones of inhibition (mm) by: *L. javanica* essential oils against *S. aureus* (100 mg/ml)

Organism	Zones of inhibition (mm)
<i>S. aureus</i> (ORSA)	13
<i>S. aureus</i>	13
Control	6

This was the highest zone of inhibition and was not varying from strain to strain. Concentrations of 20 mg/ml to 80 mg/ml had P = 0.079 in the inhibition until a threshold of 160 mg/ml was reached. *K. pneumoniae* and its strains (including multidrug resistant one) had the maximum inhibitory zone of 22 cm at 80 mg/ml. Although *S. aureus* was sensitive to the treatment at relatively low concentration, reaching the peak at 40 mg/ml, any further increase in the drug concentration did not matter. So the responses of the other strains of the organism remained constant when pooled (StDev. = 1.069). There is a clear indication that the extract has efficacy on Gram negative

bacteria with, seemingly, higher sensitivity as compared to Gram positive, *Klebsiella spp* which increased proportionally in zones of inhibition with the increase in plant extract concentration. The effect was the same with *S. aureus*. However, it was interesting to note that there was no activity against *C. albicans* although it is considered to be a weak organism. There is a possibility of the organism having been infiltrated indirectly with foreign genetic material that enables it to exhibit resistance to the drug [3].

F. saligna gave results which indicated a high activity on *P. aeruginosa* and its strains in that it was the only single plant species whose extract gave an ever-increasing zone of inhibition proportional to the increase in concentration without deviation. The inhibition was the same for both *S. aureus* and *K. pneumoniae*. Apart from the concentrations in other species of the pathogens, the same concentrations there were no inhibitions in all the strains and species of *E. coli*, *E. faecalis* and *S. typhi*.

Toddalia asiatica root bark extracts of the plant had significant activity, $P < 0.05$ with a pooled $StDev = 0.725$ and all the tested strains of *S. aureus* and *E. coli* with growth being inhibited significantly. Its inhibition increased proportionally with the increase in the drug concentration without any or much variation. Sensitivity of the organism to the plant extract justifies the fact that the community uses the plant against respiratory tract ailments such as cough, cold, stomachache and other chest related conditions. The plant was also mentioned to be used as tonic, possibly as an immune booster whereby, the roots are boiled decanted and the decoction taken by the Ogiek [4]. However, members of the Rutaceae are known to possess strong antimicrobial agents which have been tested [5]. The genera *Pseudomonas*, *Enterococcus* and *Salmonella* were all resistant to various levels of treatments of the root bark extracts.

Olinia rochetiana was indicated to be used in the treatment of colds and chest related conditions, it was found to be active against several organisms, both Gram positive and Gram negative. The results showed that the higher the concentration of the extract, the higher the inhibition with high efficacy being reached in the case of *Pseudomonas spp.* without any variations. However, *Salmonella spp.* manifested varied species and strains responses to the extract at $P < 0.45$ without any variation in the

changes of the concentrations. In the entire tests against other pathogenic organisms, the methanol extracts was active against all the other six; namely *E. faecalis*, *S. typhi*, *P. aeruginosa*, *S. aureus*, *K. pneumoniae* and *E. coli* at varied concentrations. These results are important for the selection of the plant for future anti-bacterial studies using plant extracts (Fig. 4). The plant is known to possess prusalin hence this activity is attributed to the presence of prusalin, hydrogen cyanide and a functional, benzenoid [6]. Several species from the family have been reported to be containing various substances of which *O. cymosa* has prusalin and is used for stomachache [6].

O. africana and *O. hochstetteri* (Oleaceae), both plant species gave results with significant effect at $P < 0.045$, with changes of extracts concentrations on inhibitions growth of *S. aureus* and its other strains. There was more activity, individually, in that the area the test organisms inhibited increased proportionally with the increase in the drug concentrations. *O. hochstetteri* stem bark extracts showed high inhibitory activity against species of *E. faecalis* and its other strains at higher concentrations. This was evident since the significant effect was reliable at 95% confidence level ($p < 0.045$). Besides *O. hochstetteri* which yields olive oil that is used in cooking and pharmaceutical preparations, members of the same genus are used in ethno medicine as in the case of East African communities. There are not many mentions of the other uses of the family members in traditional medicines [7].

Root bark methanol extracts, *R. keniensis* (Rosaceae) were active with varied efficacies against several microorganisms. There was significant effect $P < 0.05$ which might help give actual efficacies with change of the extracts concentrations on inhibition of the growths of all the species and strains of *Pseudomonas*. Higher concentrations of the extract was more efficient as compared to the lower ones with pooled $StDev = 0.258$. The results were the same as in the cases of *E. faecalis*, *K. pneumoniae* and *S. aureus*. However, in the entire last cases the readings were the same. Studies to ascertain the efficacy or the safety of this genus have not been done either locally or elsewhere to justify their continued use in folklore medicine.

Four of the test organisms including their strains were highly inhibited by methanol extracts of *C. abyssinica* at increasing concentrations. *S.*

aureus had the highest area of inhibition in relation to increase in concentration with a mean of 15.7 mm at a concentration of 160 mg/ml. *K. pneumoniae* and its strains also showed similar trend of inhibition. *Pseudomonas* spp and their strains also showed the same trend of inhibition with increased concentrations of the drug. Similar activities as in the case of *Pseudomonas* and *Klebsiella* were exhibited in *E. faecalis* regardless of the strains or species. The plant was, however, not regarded as medicinal but as inclusion as tonic during cookery particularly when cooking mutton could explain its continued use by the community. Root bark extracts from *C. abyssinica* plant, the test result further showed that there was a significant effect $P=0.05$. Change of the extracts concentration showed increase on inhibitions of *K. pneumoniae*. Higher concentrations of the extract inhibited more of the bacterial growth. However, there was no significant difference in concentrations- 0.04 and 0.08gm. It was noted that there was no significant difference in inhibition on the various strains of *K. pneumoniae* even though, *K. oxytoca* was more inhibited than the other strains.

E. schimperi was widely mentioned for its medicinal value, as an antibiotic and anthelmintic [8]. Uses of the stem bark as medicine were verifiable since the methanol extract was capable of inhibiting *S. aureus*, *P. aeruginosa* and *E. faecalis*, and all their different strains. During the laboratory studies, some of the organisms, in all the cases, had decreases in area of inhibition of the pathogens in relation to increase in the extracts concentration with a significant effect at $P=0.044$. The inhibition value remained the same despite varying concentrations of the drug from one organism to the other. Similarly, there are other members of the same genus, *E. ribes* and *E. tsjenam-cattam* which have been used as taenicides, treatment of chest infections, skin ailments, cholera, pneumonia and diarrhoea in Ayurvedic medicine with lots of success [8,9].

E. capensis was most effective progressively and significantly against all types of the genus *Klebsiella* and species of *S. aureus* at a peak concentration of 40 mg/ml ($P<0.05$). Various extracts from the plant have been used as antibiotics in folklore medicine by Venda communities of South Africa with reasonable success [10]. Most of the uses are against diarrhoea, stomachache and pneumonia. Such uses tally with the cases amongst the Ogiek.

G. buchannani, (Lusaciaceae family), a medium sized tree, whose wild fruits are common and a delicacy in this area, the portion from the roots is used against stomach pain in pediatrics. Its root bark methanol extract showed high inhibition on *S. aureus*, and its resistant strains. This further demonstrates the accuracy with which the community have understood and judiciously selected products of Mother Nature to meet their daily livelihoods. Another study on the same genus elsewhere was on *G. mangostana* which has been reported to possess strong antimicrobial activities against several strains of the species, *S. aureus* [11].

I tinctoria, (Balsaminaceae), was mentioned by the community to be useful in enhancing delivery process in human. The root extract from the plant when screened against the test organisms had high inhibitory efficacy on *S. aureus* and its strains. Furthermore, the extracts showed high inhibition on all the test fungi except marginally *M. mentagrophyte*. The medicinal uses of the species is not cited in literature elsewhere in the world to authenticate this report and for that matter further investigations are recommended on this plant.

L. javanica (Table 13) (Verbenaceae) and *S. biflora* (Lamiaceae) are used in fresh wounds treatment, pneumonia, and colds, and in tea as tonic respectively. Oil extracted from *S. biflora* was effective against *S. aureus*, *K. pneumoniae* and *E. coli* including all their drug resistant strains. The essential oils from *L. javanica* possessed activities against all the strains of *S. aureus*, at the levels shown in the Tables 14 and 12. *L. javanica* is amongst other medicinal essential oils that have been studied elsewhere and proved useful medically. Essential oils from higher plants have been used in folklore medicines from time immemorial, and the uses are universal in all the continents, through civilisations [12]. There are other plants that have shown activities which include the genus *Ocimum* from which oils capable of controlling *Propionibacterium acnes* is found [12]. *Thymus kotschyanus* yielded oils that were active against *Helicobacter pylori* which are found in human duodenal ulcer sites [13]. *Pseudomonas aeruginosa* which is a common opportunistic pathogen may as well be controlled by oils extracted from *Melaleuca alternifolia* L [14]. However, *S. biflora* which has varied activities when screened in the laboratory *in vitro* has not been studied exhaustively. The plant oil extracts gave promising and astounding results against

both Gram negative and Gram positive bacteria (Tables 14 and 12).

Table 14. Zones of inhibition (mm) by *S. biflora* (conc.100 mg/ml) against organisms

Pathogens	Zones of inhibition (mm)	Control (mm)
<i>S. aureus</i> (ORSA)	19	6
<i>S. aureus</i>	10	6
<i>K. pneumoniae</i>	10	6
<i>K. pneumoniae</i> (MDRS)	11	6
<i>E. coli</i>	17	6

Laboratory results indicated strong activities of essential oil extracts from *S. biflora* against *S. aureus*, *E. coli* and *K. pneumoniae* which is a clear justification of its use by the communities of freshly prepared decoction against respiratory ailments and abdominal complaints. (Tables 12 and 14) Coincidentally, a decoction from the plant is also taken as tonic. However, the use of zones of inhibition was not conclusive enough to determine the efficacy of a drug.

4. CONCLUSIONS AND RECOMMENDATIONS

The plants which did not show any bio-activities against the test pathogens could be further studied to ascertain their efficacies and the plants that showed bioactivities be studied *in vivo* using small laboratory animals to validate their efficacies. There should be a proper epidemiological record keeping so as to establish the efficacy of the drugs used in traditional systems on endemic diseases and the newly emerging and reemerging ones. The toxicity of plant medicines to human and livestock be carried out by laboratory cytological evaluations using small animal models.

More organisms should be covered in future studies so as to leave no doubt on the validation of the efficacy of such plants. For purposes of scientific progress, phytochemists should be carry out comprehensive elucidation of the active principles. Thereafter, pharmacists may work on the several combinations and cocktails which could be effective and used in the manufacture of new drugs to combat the maladies currently afflicting humankind. Further assays on these sequential extracts against some common human pathogenic fungi and profile the classes found in the biologically active plants.

Several plants that were found to contain essential oils of reasonable efficacies against known human and animal pathogens, further researches are carried on their possible incorporation into toiletries, and other oral medicines for purposes of pharmaceutical uses.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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