Journal of Biology, Agriculture and Healthcare
ISSN 2224-3208 (Paper) ISSN 2225-093X (Online)
Vol.4, No.28, 2014

Antimicrobial Activity, Toxicity and Phytochemical Screening of Four Medicinal Plants Traditionally Used in Msambweni District, Kenya

Magrate M. Kaigongi[1*] Saifuddin. F. Dossaji [1] Joseph. M. Nguta[2] Catherine.W. Lukhoba[1]
Fredrick. M. Musila[1]
1.School of Biological Sciences, College of Biological & Physical Sciences, University of Nairobi, P.O. Box 30197-00100, Nairobi, Kenya
2.Department of Public Health, Pharmacology &Toxicology, College of Agriculture &Veterinary Sciences, University of Nairobi, P.O. Box 29053-00625 Nairobi, Kenya
* E-mail of the corresponding author: kaigongim@gmail.com

Abstract
This study was designed to evaluate the antimicrobial activity, toxicity and phytochemical composition of organic and aqueous crude extracts of *Zanthoxylum chalybeum* Engl. (Rutaceae), *Adansonia digitata* L. (Bombacaceae), *Launaea cornuta* (Hocht. ex Oliv. & Hern) C.Jeffrey (Compositae) and *Grewia trichocarpa* Hochst. ex A.Rich (Tiliaceae) traditionally used by local communities of Msambweni District in Kenya. Aqueous and organic [Chloroform: Methanol (1: 1)] crude extracts were evaluated for their *in vitro* antimicrobial activity against Methicillin resistant *Staphylococcus aureus (*MRSA*)*, *Bacillus cereus, Escherichia coli, Pseudomonas aeruginosa* and *Candida albicans* using broth dilution and disc diffusion methods. Toxicity was determined using Brine-shrimp larvae (*Artemia salina* L. nauplii) assay. The crude extracts were screened to determine the presence of flavonoids, alkaloids, saponins and sesquiterpene lactones using standard techniques. It was observed that the organic crude extracts from all the species tested except *L. cornuta* exhibited dose dependent activity against *B. cereus*, MRSA, *P. aeruginosa* and *C. albicans*. None of the crude extracts showed any inhibition against *E. coli. Adansonia digitata* and *Grewia trichocarpa* had LC_{50}>1000 µg/ml and were shown to be non-toxic to Brine shrimp larvae unlike those of *Z. chalybeum* and *L. cornuta* which both had LC_{50}<500 ug/ml and were considered to be toxic. Phytochemical screening of the crude extracts showed that alkaloids, flavonoids, sesquiterpene lactones and saponins were present in the four plants tested.The study has shown that *A. digitata* and *Z. chalybeum* possess promising antimicrobial activity against microbes of health importance and could lead to the isolation of new and potentially effective antimicrobial compounds.
Keywords: Medicinal plants; Antimicrobial activity; Brine shrimp lethality test; Phytochemical analysis; Msambweni district; Kenya.

1. Introduction

A two-fold increase in microbial resistance against antibiotics has developed over the last forty years in both medical and livestock sector (Daboor and Haroon, 2013). Microbial infections have caused a big burden of diseases and bacteria are listed in the first position among common microorganisms responsible for opportunistic diseases associated with HIV/AIDS (Rathee *et al*., 2012). Increased antibiotic resistance has become a global concern, coupled with the problem of microbial persistence, thus highlighting the need to develop novel antimicrobial drugs that are not only active against drug resistant microbes, but more importantly, kill persistent micro-organisms and shorten the length of treatment (Mariita *et al*., 2010). According to World Health Organisation (2001), 80% of African and Asian populations depend on traditional medicine from plant biodiversity for primary health care because plant derived medicines are relatively cheaper and safer compared to the synthetic alternatives. Many commercial drugs used in modern medicine were derived from plants following ethno-botanical and ethno-medical knowledge (Arokiyaraj *et al*., 2012). The World Health Organisation (WHO, 2001) has advocated traditional medicine as a safe remedy for ailments of microbial and non-microbial origin. Plant derived secondary metabolites such as alkaloids, flavonoids, phenolics, saponins, and tannins protect plants from invaders such as fungi, bacteria, viruses, and nematodes. It is estimated that among the traditional medicinal plants, only 12,000 or less than10% have undergone pharmacological evaluation (Marimuthu *et al*., 2011). There is therefore a need to carry out more research on traditional medicinal plants for new antimicrobial agents as alternative to available antibiotics as they may prove to be effective against resistant pathogens to avoid the threat of post antibiotic era (Marimuthu *et al.,* 2011). Biological activity of plants is attributed to the class and concentration of phytochemical constituents which makes some plant extracts exhibit a variety of activities (Wang *et al*., 2010). Phytochemical screening of plant extracts especially those which have been used in traditional medicine is therefore essential so as to identify phytochemical constituents in the plants that are responsible for a given bioactivity. The current study investigated the antimicrobial activity, toxicity and phytochemical composition of four plants traditionally used in Msambweni ethnomedicine

2. Materials and Methods

2.1. Collection of plant materials: Various plants parts depending on the ethnopharmacological knowledge (Nguta *et al*., 2010a, b) were collected from Msambweni District in Kenya. The plants were identified by a taxonomist from the University of Nairobi and voucher specimens deposited in the Nairobi University herbarium. Voucher numbers (in parenthesis) are: *A. digitata* (JN01), *G. trichocarpa* (JN02), *L. cornuta* (JN03) and *Z. chalybeum* (JN04). The plants parts were air dried at room temperature, chopped into small pieces and ground into powder. Collected plant parts used in the study were: *A. digitata* stem bark, *G. trichocarpa* roots, *L. cornuta* leaves and *Z. chalybeum* root bark.

Preparation of crude extracts: Aqueous and organic crude extracts were prepared by cold maceration, whereby 50 grams of the powdered plant material was extracted in 500 mls of distilled water and chloroform-Methanol mixture (1:1) respectively. The plant material was exhaustively extracted over 48 hrs period. The aqueous extracts were filtered, frozen and lyophilized into a dry powder. Organic extracts were filtered and concentrated with a rotary evaporator to dry powder. The recovered extracts were stored at 4^0C for further use.

2.2. Microorganism suspension standardization: *Bacillus cereus* (ATCC11778), *Pseudomonas aeruginosa* (ATCC27853), *Escherichia coli* (ATCC25922) and *Candida albicans* (ATCC10231) were obtained from the Department of Public Health, Pharmacology and Toxicology, University of Nairobi while Methicillin Resistant *Staphylococcus aureus* (MRSA) was obtained from the Centre for Microbiology Research (CMR), Kenya Medical Research Institute (KEMRI). The fungal suspension was standardized according to the Clinical and Laboratory Standards Institute (CLSI, 2008). The yeast was grown in Sabouraud agar for 72 hours and standardized with sterile saline to turbidity equivalent to 0.5 McFarland scale (approximately $1\text{-}5\times10^6$ CFU/ml). Bacteria were grown and standardized according to (CLSI, 2009). This was done by growing them in Muller–Hinton agar for 24 hours and standardizing them with sterile saline to turbidity equivalent to 0.5 McFarland scale (approximately$1\text{-}2\times10^8$CFU/ml) and stored at 4^0C. The antibacterial and antifungal activity was determined using Agar well diffusion test and broth dilution method (Kalayou *et al*., 2012).

2.3. Disc diffusion: To determine susceptibility, four concentrations (200 mg/ml, 100 mg/ml, 50 mg/ml and 25 mg/ml of crude extracts) were prepared in dimethylsulfoxide (DMSO) and water for organic and aqueous extracts respectively. Sterile antibiotic assay discs (Whatman, 6 mm) were impregnated with 100µL of the reconstituted extract and dried completely under sterile conditions in a laminar flow. Each disc was gently pressed down to ensure complete contact with the agar inoculated with 1ml of test microorganisms (bacteria and fungi). Extracts were tested in triplicate, (Kaloyou *et al*., 2012). Dimethylsulphoxide (DMSO) and water saturated assay discs were used as negative control. Gentamicin and Amphotecirin B were used as positive control for bacteria and fungi respectively. The plates were incubated at 37 ^{0}c for 18-24 h for bacteria and 30^0c for 72 hrs for fungi. Inhibition zones were recorded as the diameter of growth free zones.

2.4. Determination of minimum inhibitory concentration (MIC): Broth dilution method was used to determine minimum inhibitory concentration (MIC). One mL of 24 h culture of test organisms (107 CFU/mL) adjusted to McFarland turbidity standard were incubated in serial dilution ranging from 10 to 200 mg/mL of plant extracts in DMSO for organic extracts and distilled water for aqueous extracts at 37^{0C} for 24 hrs for bacteria and 30^{0C} for 72hrs for fungi. The concentration of the lowest dilution with no detectable bacterial growth was considered as MIC. Absence of growth was confirmed by absence of turbidity and by inoculating into agar.

2.5. Acute toxicity testing: Toxicity of the extracts was evaluated using Brine shrimp assay (Musila *et al*. 2013). Brine Shrimp eggs were hatched in sea water for 48hrs at room temperature. Ten larvae were transferred to 5mls of 10µg/ml, 100µg/ml and 1000µg/ml of the crude extracts dissolved in sea water for aqueous extracts and DMSO for organic extracts. These were incubated at room temperature for 24hrs and live larvae were counted. DMSO and sea water were used as controls for organic and aqueous extracts respectively. The tests were done in triplicate. LC_{50} was determined by Finney's probit analysis (Finney, 1971).

2.6. Thin Layer Chromatography (TLC): Crude plant extracts were screened for flavonoids, alkaloids, sesquiterpene lactones and saponins (Harbourne, 2002).

2.7. Data analysis: Statistical analysis of antimicrobial activity was done using SPSS (statistical analysis software). ANOVA was used to determine whether there were significant differences in the mean diameter of inhibition zones in various concentrations. Dunnett test was used for multiple comparisons of inhibition. The significance level used in the analysis was 0.05 (Alpha Level $\leq$ 0.05). In toxicity testing,

3. Results

3.1. Disc diffusion technique

The results of antimicrobial screening of the extracts of test plants are shown in Table1 and Table 2. The organic crude extracts of all plants showed larger inhibition zones compared to the aqueous crude extracts. All the plants tested showed antimicrobial activity by inhibiting one or more microorganisms. Among these, *A. digitata* and *Z. chalybeum* organic extracts showed promising activity against Methicillin Resistant *S. aureus* (MRSA) and *B. cereus* respectively.

Table 1: Antimicrobial activity of aqueous and organic extracts.

Plant name	*Conc. (mg/ml)*	*C.albicans*		*E.coli*		*P.aeruginosa*		*B.cereus*		*S.aureus*	
		Aq	*Org*	*Aq*	*Org*	*Aq*	*Org*	*Aq*	*Org*	*Aq*	*Org*
A.digitata L.	200	0	9.7	0	0	9.17	9.17	8.67	8.67	9.33	13.67
	100	0	8.3	0	0	8	9	7.5	7.5	8.17	12
	50	0	0	0	0	7.5	8.5	0	0	0	9.33
	25	0	0	0	0	0	0	0	0	0	8.33
Z.chalybeum .Engl.	200	0	8.17	0	0	0	10.5	10.5	13.17	0	9.33
	100	0	7.5	0	0	0	9.17	7.67	12.5	0	9.33
	50	0	7	0	0	0	8	0	9.2	0	8.67
	25	0	0	0	0	0	7.67	0	7.00	0	0
L.cornuta (Hocht.ex.Oliv.)C.Jeffrey	200	0	0	0	0	0	10.5	0	0	0	8
	100	0	0	0	0	0	9.17	0	0	0	7.5
	50	0	0	0	0	0	8	0	0	0	7
	25	0	0	0	0	0	0	0	0	0	0
G.trichocarpa Hochst. ex A.Rich	200	0	8.5	0	0	0	10.67	0	11.67	0	10.33
	100	0	7.83	0	0	0	9.67	0	9.67	0	9.17
	50	0	0	0	0	0	8.83	0	7.17	0	8
	25	0	0	0	0	0	7.33	0	0	0	0
Gentamicin	0.04	-		14.5		15.17		15.83		15.5	
Amphotericin B	0.03	15.67		-		-		-		-	

3.2. Broth dilution technique

Majority of aqueous extracts did not show any antimicrobial activity against most of the microbes at any concentration tested and therefore their MIC values were assumed to be more than 200 mg/ml. The organic extracts of *A. digitata, Z. chalybeum* and *G. trichocarpa* had lowest MIC value of 20mg/ml against MRSA, *B. cereus* and *P. aeruginosa* respectively as shown in table 2

Table 2: Minimum Inhibitory Concentration

Plant name	Minimum inhibitory concentration (mg/ml)									
	C.albicans		*E.coli*		*P.aeruginosa*		*B.cereus*		MRSA	
	Aq	Org	Aq	Org	Aq	Org	Aq	Org	Aq	Org
A.digitata L.	> 200	70	> 200	> 200	40	30	90	90	80	20
Z.chalybeum .Engl.	> 200	40	> 200	> 200	> 200	20	90	20	> 200	40
L.cornuta (Hocht.ex.Oliv.)C.Jeffrey	> 200	> 200	> 200	> 200	> 200	40	> 200	> 200	> 200	40
G.trichocarpa Hochst. ex A.Rich	> 200	80	> 200	> 200	> 200	20	> 200	40	> 200	40

3.3. Brine-Shrimp lethality test

A. digitata aqueous crude extract had the highest LC_{50} value of 3988 followed by *G. trichocarp*a with 1488 while *Z. chalybeum* exhibited the least LC_{50} value of 293.5. *A. digitata* organic crude extract showed the highest LC_{50} value of 5626 followed by *G. trichocarpa* with 5602. *L. cornuta* showed LC_{50} value of 141.3 while *Z. chalybeum* recorded the lowest LC_{50} value of 39.2 as shown in table 3.

Table 3: Toxicity of the crude plant extracts.

Plant name	Average mortality at various concentrations (µg/ml)								LD_{50}(µg/ml)	
	1,000		100		10		0			
	Aq	Org	Aq	Org	Aq	Org	Aq	Org	Aq	Org
A.digitata L.	1.33	2.67	0.33	1	0	0	0	0	3988	5626
Z.chalybeum .Engl.	8	10	1	5.67	0	3	0	0	293.5	39.2
L.cornuta (Hocht.ex.Oliv.)C.Jeffrey	6.33	8.33	1.67	4	0.33	1.33	0	0	554.4	141.3
G.trichocarpa Hochst. ex A.Rich	0.33	2.33	0	0	0	0	0	0	1488	5602

3.4. Phytochemical screening of secondary compounds

Alkaloids and flavonoids were present in all the aqueous extracts while saponins were only found in L. cornuta only. Sesquiterpene lactones were absent in all the aqueous extracts. Alkaloids, flavonoids, Sesquiterpene lactones and saponins were present in all the organic extracts. The number of + shows the intensity of colouration while – shows absence of the compounds in the respective plants.

Table 4: Phytochemical screening of aqueous and CHCl3: MeOH crude extracts.

Plant name	Alkaloids		Flavonoids		Sesquiterpene lactones		Saponins	
	Aq	Org	Aq	Org	Aq	Org	Aq	Org
A.digitata L.	+	++++	+	+	-	+	-	++
Z.chalybeum .Engl.	++	+++	+	+++	-	+	-	+++
L.cornuta (Hocht.ex.Oliv.)C.Jeffrey	+	+	+	++	-	+++	+	++
G.trichocarpa Hochst. ex A.Rich	+	++	+	+	-	++	-	+

4. Discussion

The current study was designed to investigate the antimicrobial activity, toxicity and phytochemical screening of four medicinal plants used in Msambweni District, Kenya. From this study, it is clear that the chloroform: methanol (1:1) solvent extracts of all the plant were more potent than those of aqueous extracts against all the tested microbes. *A. digitata* organic extract was active against all the microbes except the *E. coli*. The highest activity of *A. digitata* recorded was against MRSA followed by *P. aeruginosa* and *B. cereus* while the inhibition of *C. albicans* was moderate. These results are in agreement with Staden *et al*, (2011) who reported that *A. digitata* has considerable antimicrobial activity against *Bacillus subtilis, S. aureus* and *C. albicans*. This signifies that, *A. digitata* extract has potential novel antimicrobial compounds for management of diseases caused by the above microbes.

L. cornuta aqueous extract did not show any inhibition against the tested microbes at any concentration. However, the organic extract showed moderate activity against *P. aeruginosa* and MRSA and failed to inhibit *B. cereus, E. coli* and *C. albicans*. This is in agreement with findings from Nagata *et al*. (2011) who reported that leaves and roots of *L. cornuta* are used as antimicrobial by people of Suba district-Kenya particularly those living with HIV/AIDS to cure opportunistic diseases.

Z. chalybeum showed antimicrobial activity with the highest activity being observed against *B. cereus* whereby at 200 mg/ml, there was no significant difference in terms of inhibition zones (mm) with that of standard antibiotic (Gentamicin, 0.04 mg/ml) ($P \geq 0.05$). *Z. chalybeum* organic extract also showed high activity against *P. aeruginosa* and moderate activity against MRSA and *C. albicans* but failed to inhibit the growth of *E. coli* at all concentrations. These results are in agreement with studies by Adesina's (2005) findings that the ethanol extract of *Z. chalybeum* has antibacterial activity against *B. cereus* and *P. aeruginosa* and antifungal activity against *C. albicans*.

G. trichocarpa organic extract ($CHCl_3$: MeOH) showed considerable antimicrobial activity against four of the five tested microbes; MRSA, *B. cereus, P. aeruginosa* and *C. albicans* and failed to show any inhibition against *E. coli.* The aqueous extract of *G. trichocarpa* did not inhibit any of the tested microbes. The organic extract showed high activity against *P. aeruginosa* and moderate activity against *C. albicans*, *B. cereus* and MRSA. This is the first report on antimicrobial activity of *G. trichocarpa*.

The organic extracts were more active than aqueous extracts. Noting that the traditional herbal remedy preparation from these plants is by use of water, it is a paradox that the aqueous extracts were inactive in this study. This may be due to the absence or insufficient and ineffective concentration of the antimicrobial constituents in the aqueous extracts. These findings are in agreement with the studies of Koua *et al*., (2011) who reported that inactivity of aqueous extracts of *Striga hermonthica* may be due to absence or insufficient and effective concentration of the antimicrobial agents of *S. hermonthica* extracts. It is well known that patients using the traditional herbs take in large amounts of the concoctions and hence may eventually consume sufficient

amounts of the curative drugs to elicit healing.

The study on toxicity of *A.digitata* against Brine-shrimp confirmed that both the aqueous and organic extract of *A. digitata* were non- toxic since both extracts had LC_{50} > 1000 µg/ml, an observation that is in agreement with that of Musila *et al*., (2013). *A. digitata* has also been shown to be non toxic on mice from previous studies. For instance, LD_{50} of aqueous extract of the fruit pulp of *A. digitata* on mice was found to be over 8000 µg/ml (Ramadan *et al*., 1994). This corroborates the results obtained in this study on the non toxicity of stem bark of *A. digitata* on Brine shrimp larvae. The non toxicity of *A. digitata* stem implies that the plant may be used to inhibit the growth disease causing bacteria and fungi without eliciting any toxicity on the patients. Toxicity study of *L. cornuta* on Brine shrimp from this study showed that both the aqueous and organic extracts ($CHCl_3$: MeOH) were toxic particularly the ($CHCl_3$: MeOH) ones. Similarly high levels of toxicity of stem bark of *L. cornuta* on Brine shrimp larvae has also been reported by Nguta *et al* .(2011) and Musila *et al*. (2013).

The study on toxicity of *Z. chalybeum* found that both the aqueous and organic ($CHCl_3$: MeOH) extracts of *Z. chalybeum* were highly toxic against brine shrimp larvae. Nguta *et a*l. (2011) while investigating toxicity of aqueous extracts of the leaves, stem bark and root bark of *Z. chalybeum* on Brine shrimp larvae obtained LD50<500 µg/ml for the three plant parts. This confirms the results obtained in this study and implies that *Z. chalybeum* may not make safe antimicrobial herbal remedies. This calls upon cautious use of the plant through dose adjustment amongst communities using this plant for preparation of herbal decoctions. Similarly, toxicity of *Z. chalybeum* methanolic root bark extract on human normal fetal lung fibroblast cells has also been reported by Kamuhabwa *et al*. (2000).

Toxicity study showed that both the organic and aqueous extracts of *G. trichocarpa* were non toxic to brine shrimp larvae. This is the first report on the toxicity studies of *G. trichocarpa.*

The organic extract of *G. trichocarpa* contained alkaloids, flavonoids, saponins and sesquiterpene lactones while the aqueous extract contained only alkaloids and flavonoids. There is no available report from the literature on the antimicrobial activity, toxicity and phytoconstituents of *Grewia* species except on *Grewia hexaminta* which is said to contain triterpenoid compounds (Raghunathaiyar, 1996). This could be the first time to report on *G. trichocarpa* phytochemistry.

Thin layer chromatography (TLC) study of *A. digitata* stem bark shows that the organic extract contained alkaloids, flavonoids, saponins and sesquiterpene lactones while the aqueous extract lacked both saponins and sesquiterpene lactones and contained alkaloids and flavonoids only. This is in agreement with Musila *et al*. (2013). In addition, *A. digitata* stem bark contains medicinal compounds which are largely classified under saponins, alkaloids and flavonoids such as lupeol acetate, β-sitosterol, scopoletin, friedelin, betullinic acid and adansonin while the fruit pulp of *A.digitata* is rich in procyanidins (Sidibe & Williams, 2002; Shahat, 2006). Alkaloids, flavonoids and sesquitepenes have been reported to be potent plant secondary metabolites with broad spectrum of bioactivities (Mazid *et al*., 2011). The higher antimicrobial activity of *A. digitata* on MRSA could be attributed to the presence of large amounts alkaloids. Alkaloids are pharmacogenically active basic principles of flowering plants, (Das *et al*., 2010). The observed activity of these compounds in *A. digitata* is in line with that of Karou *et al*. (2006) who demonstrated that the Indoloquinoline alkaloid causes cell lysis and morphological changes of *S. aureus.* Secondary metabolites screening in this study found that both aqueous and organic ($CHCl_3$: MeOH) extracts of *L. cornuta* contained flavonoids, alkaloids, saponins with sesquiterpene lactones lacking in the aqueous extract. These results are in agreement with (Musila *et al*., 2013). Elsewhere in Ali *et al*. (2003), the genus *Launaea* is characterized for flavonoids, triterpenes, sesquiterpene lactones, coumarins and steroids. Various species of *Launaea* such as, *L. arborescens*, *L. mucronata, L. nudicaulis* and *L. capitata* contain various types of flavones such as luteolin, apigenin and flavone glycosides such as apigenin 7-O-glucoside, vitexin, luteolin 7-O-glucoside and luteolin 7-O-rhamnoside besides others (Christian & Christian, 2010). Organic extract ($CHCl_3$: MeOH) of *Z. chalybeum* was found to contain alkaloids, flavonoids, saponins and sesquiterpene lactones while the aqueous extract contained only alkaloids and flavonoids. Similar results were obtained by Musila *et al*. (2013). *Z. chalybeum* has been reported to contain alkaloids which have antibacterial and cytotoxic activity (Chrian *et al.*, 2011. *Zanthoxylum* species contains various compounds such as alkaloids, aliphatic and aromatic amides, lignans, coumarins, sesquiterpene lactones and sterols (Cheng *et al*., 2011; He *et al*., 2002).

Biological activity is attributed to the presence of various secondary metabolites in plants (Mazid *et al*., 2011). Not only their presence, but also the quantity of the phytochemical constituents in a given plant extract determines the extent of extracts' bioactivity. In addition, presence of more than one class of secondary metabolites in a given plant extract will also determine the nature and extent of extract's biological activity (Wang *et al*., 2010). The presence of flavonoids in the crude extracts facilitated the antimicrobial activity as flavonoids are known to complex with the extracellular and soluble proteins and bacterial cell wall of bacteria. More flavonoids may also disrupt microbial membrane. The sesquiterpene lactones are known to be active against bacteria, protozoa and viruses through membrane disruption. (Cowan, 1999).

5. Conclusion

The current study shows that *Z. chalybeum* and *A. digitata* inhibit the growth of *B. cereus* and MRSA respectively with no significance difference (P≥0.05) with that of standard antibiotic (Gentamicin). The crude extracts of *A. digitata* and *G. trichocarpa* proved to be non toxic on Brine shrimp larvae with LC_{50}>1000 µg/ml while *Z. chalybeum* and *L. cornuta* were found to be highly toxic with LC_{50}<500 ug/ml. Flavonoids, alkaloids, saponins and sesquiterpene lactones were present larger amounts in organic extracts of *A. digitata*, *G. trichocarpa*, *L. cornuta* and *Z. chalybeum* compared to their aqueous extracts. Alkaloids were present in high amount while sesquiterpene lactones were present in low amounts in all the extracts. The antimicrobial activity, toxicity and phytochemistry of crude extracts *G. trichocarpa* is being reported for the first time. This could be a start point for further drug research of this species on a wide diversity of microbial pathogens.

Acknowledgement

This study received partial funding from Masinde Muliro University of Science and Technology (MMUST) and DAAD. We are also grateful to the University of Nairobi for offering space and facilities to carry out this study.

References

Adesina, S.K. (2005). The Nigerian *Zanthoxylum*; chemical and biological values. *Afrian Journal of Traditional and Complimentary medicine*, **2 (**3) ,282 – 301

Ali, D., Hussein, S.M.S., Malik, A., Ahmed, Z. (2003). Chemical constituents of the genus *Launaea. Journal of Chemical Society of Pakistan*, **25**, 341-347

Arokiyaraj, S., Sripriya, N., Bhagya, R., Radhika, B., Prameela, L., Udayaprakash, N.K. (2012). High rates of multidrug resistance in Enterococcus faecalis and E. faecium isolatedfrom inpatients and outpatients in Taiwan. *Journal of Diagnostic Microbiology and Infectious Disease* **75**, 406–411

Cheng, F. W., Kai, Y., Hai, M.Z., Jie, C., Rui, F., Zhi, L. L., Shu, S.D., Yong. Y.W., Zhi, W.D., Ligang. Z. (2011). Components and Insecticidal Activity against the Maize Weevils of *Zanthoxylum schinifolium* Fruits and Leaves, *Molecules* **16**, 3077-3088.

Christian, Z & Vipaporn, S., (2010). Flavonoids as chemosystematics markers in the tribe Cichorieae of the Asteraceae. *Journal of Biochemical Systematics and Ecology* **38**, 935–957.

Clinical and Laboratory Standards Institute (CLSI), (2008). Reference Method for Broth Dilution Antifungal Susceptibility Testing of Yeasts: Approved Standard M27-A3, NCCLS, Wayne, PA.

Clinical and Laboratory Standards Institute (CLSI),(2009). Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically: Approved Standard, eighth ed. M07-A8. CLSI, Wayne, PA.

Das, K., Tiwari, R.K.S., & Shrivastava, D.K., (2010). Techniques for Evaluation of Medicinal Plant Products as Antimicrobial Agent: Current Methods and Future Trends. *Journal of Medicinal Plants Research*, **4**(2) 104-111.

Finney D.J., (1971). Probit analysis, 3rd Ed, Cambridge University press.

Harborne, J.B., (2002). Phytochemical Methods, A Guide to modern techniques of plant analysis, 3rd Ed. Chapman & Hall. London

He,W., Van Puyvelde,L., De Kimpe,N.,Verbruggen,L., Anthonissen,K.,Van der Flaas,M.,Bosselaers,J.,Mathenge,S.M.,Mudida,F.P.(2002) Chemical Constituents and Biological Activities of *Zanthoxylum usambarense*. *Phytotherapy Research*, **16**, 66–70.

Kalayou, S., Haileselassie, M., Gebre-egziabher, G., Tiku', T., Sahle, S.,Taddele, H., Ghezu, M. (2012). In-vitro antimicrobial activity screening of some ethnoveterinary medicinal plants traditionally used against mastitis, wound and gastrointestinal tract complication in Tigray Region, Ethiopia. *Asian Pacific Journal of Tropical Biomedicine* 512-522

Kamuhabwa,A., Nshimo,C.,Witte,P. (2000),Cytotoxicity of some medicinal plant extracts used in Tanzanian traditional medicine, *Journal of Ethnopharmacology* **70**, 143–149

Karou, D., Savadogo, A., Canini, A., Yameogo, S., Montesano, C., Simpore, J., ... Traore, A. (2006). Antibacterial activity of alkaloids from *Sida acuta*. *African Journal of Biotechnology*, **5**(2), 195-200.

Marimuthu, J., Mithraja, M.J.,Mony Mahesh, M., Paul, Z. M., Jeeva, S., (2011). Phytochemical studies on *Azolla pinnata* R. Br., *Marsilea minuta* L. and *Salvinia molesta* Mitch. *Asian Pacific Journal of Tropical Biomedicine* S26-S29

Masakazu, N., Firth, N.A., Cannon, D.R. (2012). Antifungal drug resistance of oral fungi. *Odontology*, **98**(1), 15-25.

Mazid, M. , Khan, T.A., Mohamad, F., (2011). Role of Secondary Metabolites in Defence Mechanism in Plants. *Journal of Biology and Medicine* **3**(2), 232-249

Musila, M.F, Dossaji, S.F., Nguta, J.M., Lukhoba, C.W., Munyao, J.M. (2013). *In vivo* antimalarial activity, toxicity and phytochemical screening of selected antimalarial plants. *Journal of Ethnopharmacology,* 146, 557-561

Nagata, J.M., Alan, R. J., John, .K. Charles,R.S.,Elizabeth,A.B.,Craig,R.C. (2011). Medical pluralism on Mfangano Island: Use of medicinal plants among persons living with HIV/AIDS in Suba District, Kenya. *Journal of Ethnopharmacology,* 135, 501–509

Nguta,J.M., Mbaria,J.M., Gakuya,D.W.,Gathumbi,P.K.,Kabasa,J.D.,Kiama,S.G. (2011). Biological Screening of Kenya Medicial Plants using *Artemia salina* L. (Artemiidae), *Pharmacology* online, 2,458-478

Ramadan, A., Harraz, F.M., EL-Mougy, S.A.,(1994). Anti-inflamatory, analgesic and antipyretic effects of the fruit pulp of *Adansonia digitata*, *Fitoterapia* 65(5), 418-422

Rathee, D., Rathee ,D., Rathee, P., Rathee, S. (2012). Phytochemical screening and antimicrobial activity of Picrorrhiza kurroa, an Indian traditional plant used to treat chronic diarrhea. *Arabian Journal of Chemistry*, in press

Shahat, A.A.,2006, Procyanidins from *Adansonia digitata*. *Journal of Pharmaceutical Biology* 44(6), 445–450

Sidibe, M., William, J.T. (2002). BAOBAB: *Adansonia digitata* L., International Center for Underutilized Crops, RPM Reprographics Chichester, England.

Wang,Y.F.,Ni,Z.Y.,Dong,M.,Cong,B.,Shi,W.Q.,Gu,Y.C., Kiyota,H. (2010). Secondary Metabolites of Plants from the Genus Saussurea; Chemistry and Activity. *Chemistry and Biodiversity* 7, 2623-2659

Wanyoike, G.N., Chabra, S.C., Lang'at, C.C., Omar, S.A., (2004). Brine shrimp toxicity and antiplasmodial activity of five Kenyan medicinal plants. *Journal of Ethnopharmacology* 90, 129–133

World Health Organization. 2001. Legal status of traditional medicine and complementary/alternative medicine: a worldwide review. Available from: http://libdoc.who.int/hq/2001/WHO_EDM_TRM_2001.2.pdf

The IISTE is a pioneer in the Open-Access hosting service and academic event management. The aim of the firm is Accelerating Global Knowledge Sharing.

More information about the firm can be found on the homepage:
http://www.iiste.org

CALL FOR JOURNAL PAPERS

There are more than 30 peer-reviewed academic journals hosted under the hosting platform.

Prospective authors of journals can find the submission instruction on the following page: http://www.iiste.org/journals/ All the journals articles are available online to the readers all over the world without financial, legal, or technical barriers other than those inseparable from gaining access to the internet itself. Paper version of the journals is also available upon request of readers and authors.

MORE RESOURCES

Book publication information: http://www.iiste.org/book/

Academic conference: http://www.iiste.org/conference/upcoming-conferences-call-for-paper/

IISTE Knowledge Sharing Partners

EBSCO, Index Copernicus, Ulrich's Periodicals Directory, JournalTOCS, PKP Open Archives Harvester, Bielefeld Academic Search Engine, Elektronische Zeitschriftenbibliothek EZB, Open J-Gate, OCLC WorldCat, Universe Digtial Library , NewJour, Google Scholar