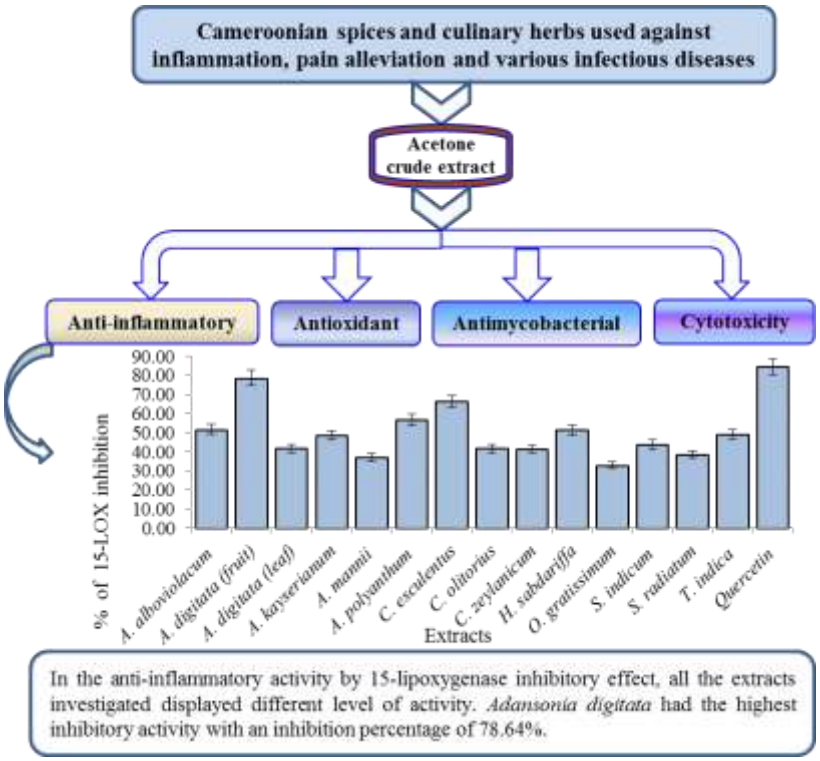
The 15-lipoxygenase inhibitory, antioxidant, antimycobacterial activity and cytotoxicity of fourteen ethnomedically used African spices and culinary herbs

Jean Paul Dzoyem[a,b], Victor Kuete[b], Lyndy J. McGaw[a], Jacobus N. Eloff[a*]

[a] *Phytomedicine Programme, Department of Paraclinical Sciences, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, Onderstepoort 0110, Pretoria, South Africa.*

[b] *Department of Biochemistry, Faculty of Science, University of Dschang, P.O. Box 67, Dschang, Cameroon.*

***Corresponding author**. Tel.: +27 125298244; fax: +27 5298304. E-mail address: kobus.eloff@up.ac.za (J.N. Eloff).*



Graphical abstract

Abstract

Ethnopharmacological relevance

Culinary herbs and spices are widely used ethnomedically across Africa. They are traditionally employed in the treatment of several ailments including inflammation disorders, pain alleviation and infectious diseases. Pharmacological studies are necessary to provide a scientific basis to substantiate their traditional use and safety. In this study, the 15-lipoxygenase inhibitory, antioxidant, antimycobacterial and the cytotoxic activities, total phenolic and flavonoid contents of fourteen edible plants were investigated.

Materials and Methods

The 15-lipoxygenase inhibitory activity was evaluated by the ferrous oxidation-xylenol orange (FOX) assay method. The antioxidant activity was determined using free-radical scavenging assays. The antimycobacterial activity was determined by a broth microdilution method against three species of mycobacteria: *Mycobacterium smegmatis*, *Mycobacterium aurum* and *Mycobacterium fortuitum* using tetrazolium violet as growth indicator. The cytotoxicity was assessed by the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay on Vero monkey kidney cells.

Results

All the extracts tested had some 15-lipoxygenase inhibitory activity ranging from 32.9-78.64%. *Adansonia digitata* (fruit) had the highest antioxidant capacity (IC_{50} values of 8.15 µg/mL and 9.16 µg/mL in the DPPH and ABTS assays respectively; TEAC of 0.75 in the FRAP assay) along with the highest amount of total phenolics (237.68 mg GAE/g) and total flavonoids (16.14 mg QE/g). There were good correlations between DPPH and ABTS values (R^2 0.98) and between total phenolics and total flavonoids (R^2 0.94). *Tamarindus indica* had significant antimycobacterial activity against *M. aurum* (MIC 78 µg/mL). As could be expected with edible plants, all the extracts had a relatively low cytotoxicity with LC_{50} values higher than 102 µg/mL with the exception of the two *Aframomum* species (33 and 74 µg/mL).

Conclusions

This study provides scientific support for some of the the traditional uses and the pharmacological activities of the culinary herbs and spices investigated. The results suggest that increasing intake of some of these herbs may be useful in preventing or reducing the progression of lifestyle-related diseases. The diversity of the pharmacological activities of the extract from the fruit of *Adansonia digitata* suggested that this plant might be valuable for application in human and animal health.

Key words: culinary herbs, lipoxygenase, antioxidant, antimycobacterial, cytotoxicity

1. Introduction

Medicinal plants have been used as medicine, as food or in food preparations to improve the flavour and taste throughout human history. Culinary herbs and spices are reported to contain bioactive compounds imparting various pharmacological properties to the food including antimicrobial, anti-inflammatory and antioxidant properties (Table 1). In this regard, the human diet has an important role in protection against oxidative stress, with the health-protecting factor being attributed to compounds with antioxidant capacity. During the last decade, many reports focused on the use of several spices and culinary herbs for improving the healthy life style of humans and suggested that the health benefits provided by these plants are attributable to the presence of various bioactive compounds (Becker, 1983). Phenolic compounds mainly flavonoids may be responsible for various bioactivities (Wijekoon et al., 2013). Natural antioxidants, predominantly phenolic compounds, are also considered to be important complementary factors for anti-oxidative stress (Rice-Evans et al., 1997) and several studies have shown that the antioxidant properties of plants could be correlated with oxidative stress defense (Costantino et al., 1992).

Table 1: Characteristics of the fourteen Cameroonian culinary herbs studied.

Plant name (Family) Voucher number	Common names	Synonyms	Part use	Ethno-medicinal use	Part use in this work	Previous Pharmacological activities investigated
Adansonia digitata (Bombacaceae) 42417/HNC	Baobab, cream-of-tartar tree, guinea tamarind, lemonade tree, monkey bread tree, sour gourd, upside-down tree	*Adansonia bahobab*, *A. baobab*, *A. integrifolia*, *A. situla*, *A. sulcata*, *A. somalensis*, *A. sphaerocarpa*, *Baobabus digitata*, *Ophelus sitularis*	Pulps, fruits, leaves, pip, bark	Analgesic, smallpox, rubella, antipyretic, astringent, immuno-stimulant anti-small pox, pains, anti-rubella, hyposensitive , kidney and bladder diseases, respiratory diseases, general fatigue, diarrhoea, insect bites, and guinea worm (Al-Qarawi et al., 2003; Becker, 1983; Kabore et al., 2011; Tanko et al., 2008; Wickens, 1979)	Leaves, fruits	Anti-inflammatory, antidysentery, antioxidant, analgesic, antidiarrheal, antimicrobial, antimalarial, antiiflammatory (Kabore et al., 2011; Mulaudzi et al., 2013; Samie et al., 2012)
Aframomum alboviolaceum Ridley K. schum (Zingiberaceae) 34888/HNC	Large amomum	*Aframomum latifolium*, *A. stipulatum*	Roots, Fruits	Spices, diuretic, anthelmintic, fever, antiparasitic, stomach troubles; diarrhoea, vermifuges (Abreu and Noronha, 1997; Neuwinger, 2000)	Fruits	Antimicrobial, antitumor and antileishmania, antidrepanocytary, Anthelmintic (Abreu et al., 1999; Mpiana et al., 2007)
Aframomum kayserianum K.schum (Zingiberaceae) 18884/SRFC	/	*Amomum kayserianum*	Fruits	Spices, anti-mumps, diarrhoea, dysmenorrhoeas, vermifuge (Tane et al., 2005).	Fruits	Antibacterial (Seukep et al., 2013)
Aframomum polyanthum K.schum (Zingiberaceae) 3981/SRFK	/	*Amomum polyanthum*	Fruits	Spices (Ayafor et al., 1994; Djeussi et al., 2013)	Fruits	Antibacterial (Ayafor et al., 1994; Djeussi et al., 2013)
Anonidium mannii (Oliv) Eng et Diels (Amonaceae) 1918/SRFK	Junglesop, soursop	*Anonidium friesianum*	Stem, bark, leaves	Spider and snake bites, bronchitis, syphilis, diarrhea, malaria, dysenteria, gastroenteritis (Betti, 2004; Kuete et al., 2013; Muganza et al., 2012; Noumi, 2011; Thomas et al, 2003)	Leaves	Antibacterial, antiprotozoal (Djeussi et al., 2013; Muganza et al., 2012)
Cinnamomum zeylanicum Blume (Lauraceae) 22309/SRFC	Cinnamon, ceylon, true cinnamon	*Cinnamomum verum*	Bark	Spices, respiratory, digestive and gynaecological ailments (Ranasinghe et al., 2012)	bark	anti-inflammatory, antioxidant, antibacterial, collagenase, urease inhibitory (Bujnakova et al., 2013; Hong et al., 2012; Seukep et al., 2013).
Corchorus olitorius (Tiliaceae) 14725/SRFC	Indian jute, Jew's mallow.	*Corchorus catharticus*, *C. quinquelocularis*, *C. malchairii*, *C. longicarpus*	Stem, leaves	Gonorrhoea, cystitis chronic, analgesic, febrifuge, antitumor, anti-inflammatory, diuretic, cardiotonic (Ilhan et al., 2007; Zakaria et al., 2006)	Whole plant	Antibacterial, antioxidant, anti-inflammatory, (Barku et al., 2013; Handoussa et al., 2013; Seukep et al., 2013).

Cyperus esculentus (Cyperaceae) 14977/SRFC	Tiger nut, yellow nutsedge, earth almond, earth nut	*C. aureus, Pycreus esculentus, C. esculentus, C. melinorhizus,C. tuberosus,*	Fruits, tubers	Respiratory infections and some stomach illnesses, cough, reast lumps and colon cancer, constipation, edible. (Bamishaiye and Bamishaiye, 2011; Ayafor et al., 1994; Djeussi et al., 2013).	Fruits	Antioxidant, antibacterial, anti-inflammatory (Biradar et al., 2010; Jing et al., 2013; Seukep et al., 2013).
Hibiscus sabdariffa (Malvaceae) 1918/SRFK	Roselle, sorrel, indian sorrel, red sorrel	*H. gossypifolius, H. acetosus, H. roselle, H. fratemus, H. digitatus, H. sanguineus, Sabdariffa rubra, S. digitata*	Flowers, leaves	Inflammatory conditions, diuretic, stomachic, laxative, aphrodisiac, antiseptic, astringent, sedative, hypertension and other cardiac diseases (Lin et al, 2001; Olaleye, 2007)	Leaves	Antibacterial , α-glucosidase and α-amylase inhibitory, antidiabetic (Djeussi et al., 2013; Salehi et al., 2013; Saxena et al., 2011).
Ocimum gratissimum (Lamiaceae) 42738/HNC	African basil, holy basil	*Geniosporum discolor, Ocimum anosurum, O. arborescens, O. viride, O. suave. O. robustum*	Leaves, roots, bud	Spices, respiratory tracts diseases, diarrhea, anti-hypertensive, malaria (Bruneton, 1999)	Roots	Antibacterial, spermatotoxicity, cytotoxicity, genotoxicity, insecticidal, antioxidant, (Akinboro and Bakare, 2013; Djeussi et al., 2013; Koubala et al., 2013; Oriakhi et al., 2014).
Sesamum indicum L. (Pedaliaceae) 42898/HNC	Sesame, benne, beny	*Sesamum orientale, S. mulayanum*	Fruits	Liniment, laxative, cough, emollient, antitumor hepatoprotective, hypoglycemic, anticonstipation, anti-carries (Awobajo et al., 2009; Munish et al., 2011; Xu and Ping, 2010)	Fruits	Wound healing, antibacterial, antioxidant (Bopitiya and Madhujith, 2013; Seukep et al., 2013; Sharif et al., 2013).
Sesamum radiatum Schum et Thom (Pedaliaceae) 8797/SRFC	black sesame, black beniseed, wild beniseed,	*Sesamopteris radiata*	Stem, leaves, seeds	Anti-cold, anticatarrate, against ocular pains and cutaneous eruptions, respiratory ailments ((Bankole et al., 2007).	Seeds	Hypoglycaemia, antimicrobial (Bankole et al., 2007; Seukep et al., 2013).
Tamarindus indica L (Caesalpiniaceae) 26326/SRFC	Indian date, madeira mahogany, tamarind tree	*Tamarindus occidentalis, T. officinalis, T. umbrosa.*	Fruits bark	Fever, gastriculcer, diarrhea, jaundice, conjunctivitis, hemorrhoid, astringent, asthma, eye inflammation (Escalona-Arranz et al., 2010; Morton, 1987).	Fruits	Antibacterial, analgesic, antihyperglycaemic (Beena et al., 2013; Djeussi et al., 2013)

/: not recorded

15-Lipoxygenase (15-LOX) belongs to the class of iron containing lipoxygenases that catalyze the incorporation of dioxygen into unsaturated fatty acid (Feussner and Wasternack, 2002). Lipoxygenases are the key enzymes in the biosynthesis of leukotrienes that play an important role in several inflammatory diseases (Funk, 2006). Inflammation is one of the manifestations of oxidative stress, and the pathways that generate the mediators of inflammation, such as adhesion molecules and interleukins, are all induced by oxidative stress (Sommer, 2005). Inhibition of LOX may influence the inflammation processes and thus be of interest for modulation of the lipoxygenase pathway. Therefore, inhibitors of oxidative stress and LOX have been considered as therapeutically useful in the treatment of many related diseases (Mehta et al., 2006). In view of the ethnomedicinal role of culinary herbs and the understanding of their role of oxidative stress in pathogenesis of multiple diseases the antioxidant and antimicrobial activity of herbal products has been investigated(Katalinic et al., 2013).

The fourteen culinary herbs and spices examined in this study are traditionally used for the treatment of a wide range of diseases and in some cases biological activities have been determined (Table 1). Although many studies have investigated the possible health effects of single phytochemicals originating from herbs or spices, only a few culinary herbs have been relatively extensively studied in terms of possible health effects. There is limited literature on the pharmacological effects of extracts of dietary herbs. Edible plants and their constituents are generally recognized to be safe, because of their traditional use without any documented detrimental impact, nevertheless scientific rationale for the safety of usage is needed. This study was conducted with the aim of determining the 15-lipoxygenase inhibitory, antioxidant, antimycobacterial and cytotoxic activities, total phenolic and total flavonoid contents of fourteen culinary herbs.

2. Materials and Methods

2.1. Plant material and extraction

The plants were purchased from local markets in the western region of Cameroon and identified at the Cameroon National Herbarium where voucher specimens were deposited under different reference numbers (Table 1). The collected plant material was dried at room temperature and ground. The powder obtained was macerated in acetone for 48 h and filtered through Whatman No1 filter paper. The filtrate obtained was concentrated under reduced pressure using a rotary evaporator to obtain the crude extract. The crude extracts were kept at 4°C until use.

2.2. Chemicals

Sodium carbonate was obtained from Holpro Analytic, South Africa. Gentamicin was purchased from Virbac, South Africa. Fetal calf serum (FCS) and minimum essential medium (MEM with L-glutamine) was provided by Highveld Biological, Johannesburg, South Africa. Phosphate buffered saline (PBS) and trypsin were purchased from Whitehead Scientific, South Africa. Doxorubicin was obtained from Pfizer. Quercetin, 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid) diammonium salt (ABTS), 2,2-diphenyl-1-picrylhydrazyl (DPPH), 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT), *p*-iodonitrotetrazolium violet (INT), Folin-Ciocalteu reagent, gallic acid, 2,5,7,8-tetramethylchroman carboxylic acid (Trolox) and potassium persulfate were purchased from Sigma-Aldrich St. Louis, MO, USA . Sodium dodecyl sulphate, potassium ferric cyanide, iron (II) sulfate and 15-lipoxygenase from *Glycine max* were provided by Sigma, Germany. Tris(hydroxymethyl)aminomethane was purchased from Sigma, Switzerland. Ferric chloride and linoleic acid were purchased from Merck,

Darmstadt and Schuchardt, Germany respectively, Xylenol orange was obtained from Searle Company, England.

2.3. Soybean lipoxygenase inhibition assay

The assay was performed according to previously described procedure (Pinto et al., 2007) with slight modifications. The assay is based on the formation of the complex $Fe3^{+}$/xylenol orange with absorption at 560 nm. 15-Lipoxygenase from *Glycine max* was incubated with extracts or standard inhibitor at 25°C for 5 min. Then linoleic acid (final concentration, 140 µM) in Tris-HCl buffer (50 mM, pH 7.4) was added and the mixture was incubated at 25°C for 20 min in the dark. The assay was terminated by the addition of 100 µL of FOX reagent [sulfuric acid (30 mM), xylenol orange (100 µM), iron (II) sulfate (100 µM), methanol/water (9:1)]. The lipoxygenase inhibitory activity was evaluated by calculating the percentage of the inhibition of hydroperoxide production from the changes in absorbance values at 560 nm after 30 min at 25°C. % inhibition = [(Absorbance of control – Absorbance of test sample)/Absorbance control] x100.

2.4. Antioxidant activity

2.4.1. Total phenolic content (TPC) determination

The total phenolic content of extracts was determined colorimetrically using a 96-well microplate Folin-Ciocalteu assay developed by Zhang et al. (2006). The total phenolic content was calculated from the linear equation of a standard curve prepared with gallic acid, and expressed as gallic acid equivalent (GAE) per g of extract.

2.4.2. Total flavonoids content (TFC) determination

Total flavonoid content was determined using the method of (Ordonez et al., 2006). A volume of 0.5 mL of 2% $AlCl_3$ ethanol solution was added to 0.5 mL of sample solution (1 mg/mL). After one hour at room temperature, the absorbance was measured at 420 nm. A yellow color is indicative of the presence of flavonoids. Total flavonoid content was calculated and expressed as mg quercetin equivalent/g of crude extract using a standard curve prepared with quercetin.

2.4.3. ABTS radical assay

The ABTS radical scavenging capacity of the samples was measured with modifications of the 96-well microtitre plate method described by Re et al. (1999). Trolox and ascorbic acid were used as positive controls, methanol as negative control and extract without ABTS as blank. The percentage of ABTS•+ inhibition was calculated using the formula: Scavenging capacity (%) = 100 - [(absorbance of sample - absorbance of sample blank) × 100/ (absorbance of control) – (absorbance of control blank)]. The IC_{50} values were calculated from the graph plotted as inhibition percentage against the concentration.

2.4.4. DPPH assay

The DPPH radical-scavenging activity was determined using the method proposed by Brand-Williams (1995). Ascorbic acid and trolox were used as positive controls, methanol as negative control and extract without DPPH as blank. Results were expressed as percentage reduction of the initial DPPH absorption in relation to the control. The concentration of extract that reduced the DPPH colour by 50% (IC_{50}) was determined as for ABTS•+.

2.4.5. Ferric Reducing Antioxidant Power (FRAP) assay

The FRAP of samples was determined by direct reduction of potassium ferricyanide ($K_3Fe(CN)_6$) to potassium ferrocyanide ($K_4Fe(CN)_6$) (electron transfer process from the antioxidant). The increase in absorbance from the formation of Pearl's Prussian blue complex following the addition of excess ferric ion was measured as described by Berker et al. (2007) with some modification. The reaction medium (210 µL) containing 40µL of the test samples or positive controls (trolox and ascorbic acid; concentration range between: 15.62 – 2000 µg/mL); 100µL of 1.0M hydrochloric acid; 20µL of 1% (w/v) of SDS; 30µL of 1% (w/v) of potassium ferricyanide, was incubated for 20min at 50°C, then cooled to room temperature. Finally, 20µL of 0.1% (w/v) of ferric chloride was added. The absorbance at 750 nm was read and blank absorbance was taken by preparing the reaction medium the same way without the addition of ferric chloride. The TEAC (Trolox Equivalent Antioxidant capacity) was calculated by dividing the slope of each sample (slope obtained from the line of best fit of the absorbance against concentration using the linear regression curve) by that of trolox.

2.5. Antimycobacterial activity assay

2.5.1. Microorganism culture

Mycobacterium smegmatis (ATCC 1441), *Mycobacterium aurum* (NCTC 10437) and *Mycobacterium fortuitum* (ATCC 6841) were cultured as described by McGaw et al. (2008). They were maintained on Middlebrook 7H10 agar slants, supplemented with glycerol or tween 20. Inocula suspensions were prepared by mixing a few microbial colonies with sterile distilled water. The suspension was diluted with sterile water to render a concentration of cells equal to standard McFarland 1 standard solution (approximately $4x10^7$ cfu/mL), and then diluted with

freshly prepared Middlebrook 7H9 broth supplemented with 10% oleic albumin dextrose catalase (OADC) to obtain a final inoculum density of approximately $4x10^5$ cfu/mL.

2.5.2. Determination of minimum inhibitory and bactericidal concentration (MIC and MBC)

The broth microdilution technique using 96-well micro-plates, as described by Eloff (1998) was used to obtain the MIC and MBC values of essential oil samples. Extracts (100 µL) at an initial concentration of 10 mg/mL were serially diluted, two-fold in 96-well microtitre plates, with equal volumes of Middlebrook 7H9 broth. Then, 100µL of inocula were added to each well to give a final concentration range of 2.5-0.019 mg/mL. The plates were incubated overnight for *M. smegmatis* and 3 days for *M. aurum* and *M. fortuitum* at 37°C. To indicate bacterial growth, 40µL of 0.2 mg/mL INT was added to each well after incubation and the plates incubated further at 37°C for 1 h. The MIC was defined as the lowest concentration that inhibited the colour change of INT (yellow to purple). The experiment was performed in triplicate. In addition, the total activity was calculated as the total mass (mg) of the extract divided by the MIC value (mg/mL). The total activity value indicates the volume to which the extract derived from 1 g of plant material can be diluted and still inhibit the growth of the microorganism (Eloff, 2000).

2.6. Cytotoxic activity

The cytotoxicity of the extracts (dissolved in acetone) against Vero monkey kidney cells was assessed by the MTT reduction assay as previously described by Mosmann (1983) with slight modifications. Cells were seeded at a density of 1 x 10^5 cells/ml (100 µl) in 96-well microtitre plates and incubated at 37^oC and 5% CO_2 in a humidified environment. After 24h incubation,

extracts (100 µl) at varying final concentrations were added to the wells containing cells. Doxorubicin was used as a positive reference. A suitable blank control with equivalent concentrations of acetone was also included and the plates were further incubated for 48h in a CO_2 incubator. Thereafter, the medium in each well was aspirated from the cells, which were then washed with PBS, and finally fresh medium (200 µl) was added to each well. Then, 30 µl of MTT (5 mg/ml in PBS) was added to each well and the plates were incubated at 37°C for 4h. The medium was aspirated from the wells and DMSO was added to solubilize the formed formazan crystals. The absorbance was measured on a BioTek Synergy microplate reader at 570 nm. The percentage of cell growth inhibition was calculated based on a comparison with untreated cells. The selectivity index values were calculated by dividing cytotoxicity LC_{50} values by the MIC values in the same units ($SI = LC_{50}/MIC$).

2.7. Statistical analysis

All experiments were conducted in triplicate and values expressed as mean ± standard deviation. Statistical analysis was performed using one way ANOVA and results were compared using the Fisher's least significant difference (LSD) at a 5% significance level.

3. Results and Discussion

Table 1 shows an overview of the spices and culinary herbs used, common names and synonyms as well as their ethno-medicinal use and their other pharmacological potential previously investigated (Table 1).

3.1. 15-lipoxygenase inhibitory activity

In this study, we used the ferrous oxidation-xylenol orange (FOX) assay method for testing extracts for 15-lipoxygenase inhibitory activity. The results presented in Figure 1 show that all

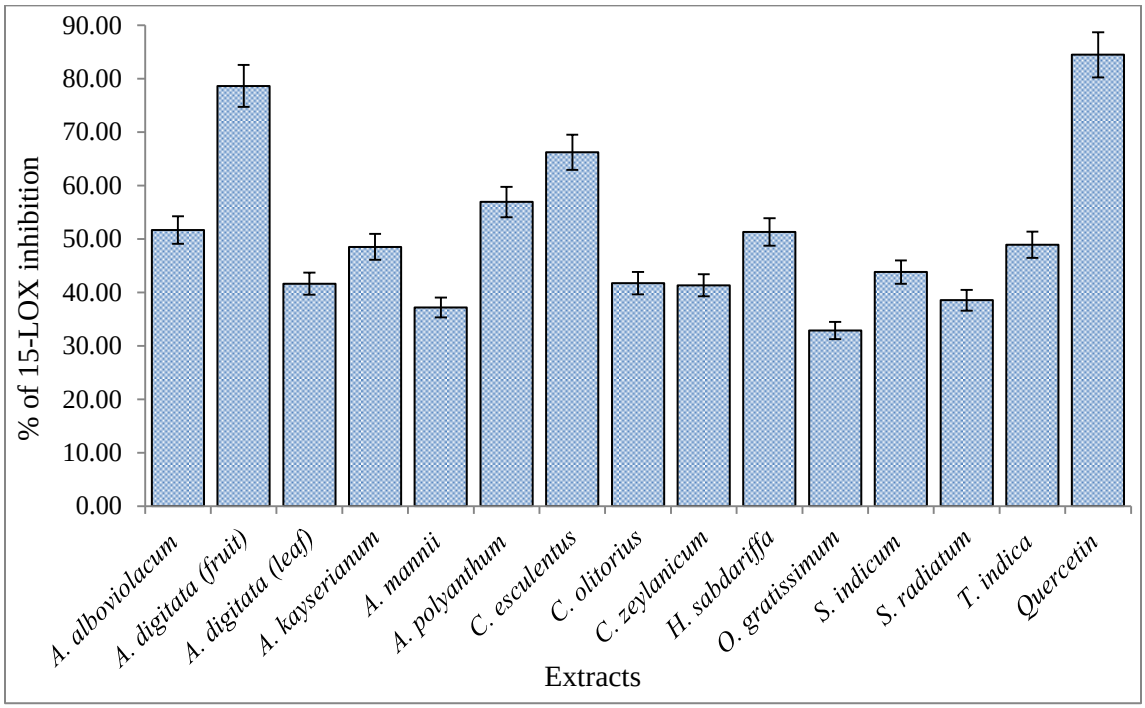


Figure 1: 15-Lipoxygenase inhibitory activity of extracts from fourteen culinary herbs. Extracts were tested at 100 µg/ml.

the extracts investigated had a certain level of 15-lipoxygenase inhibitory effect. Five extracts (*A. alboviolaceum*, *A. digitata* (fruit), *A. polyanthum*, *C. esculentus* and *H. sabdariffa*) out of fourteen led to more than 50% of 15-lipoxygenase inhibition. The observed variability in the degree of inhibition of 15-LOX by different extracts could be attributed to the differences in their phytochemical composition as previously reported by Djeussi et al. (2013). *Adansonia digitata* had the highest inhibitory activity with an inhibition percentage of 78.64%. It should be noted that *A. digitata* has been used traditionally as an analgesic and to alleviate pain (Table 1). This plant could be regarded as a promising anti-inflammatory agent. *C. esculentus* had an inhibitory

activity of 66.20%. Anti-inflammatory activity has been previously reported in *C. esculentus* (Biradar et al., 2010) as well as in *C. olitorius* and *C. zeylanicum* (Table 1). The *in vitro* lipoxygenase effect of other plant species in this study is reported for the first time. The lipoxygenase products constitute an important class of inflammatory mediators in various inflammatory diseases (Carter et al., 1991), therefore, inhibition of the biosynthesis of inflammatory mediators by blocking the activities of these enzymes would be important for the treatment of many inflammatory disease states (Benrezzouk et al., 1999).

It is noteworthy that, *A. digitata* had the highest TPC with good antioxidant activity, a finding which is consistent with Handoussa et al. (2013) who found a relationship between the anti-inflammatory activity and the presence of polyphenols. Antioxidants are also known to inhibit plant lipoxygenases (Lin et al., 2001). Studies have implicated oxygen free radicals in the process of inflammation and phenolic compounds may block the cascade process of arachidonic acid metabolism by inhibiting lipoxygenase activity, and may serve as a scavenger of reactive free radicals which are produced during arachidonic acid metabolism (Trouillas et al., 2003).

3.2. DPPH, ABTS, FRAP, Total phenolics (TPC) flavonoids content (TFC)

The free radical scavenging ability has been determined by using several different assays (Table 2). There was a very good correlation between DPPH and ABTS values (R^2 0.977) and between TPC and TFC (R^2 0.936) of the different extracts. All extracts had good antioxidant activity. Results varied depending on the method used. IC_{50} values ranged from 8.15 µg/mL to 484.86 µg/mL in the DPPH assay; and from 9.16 µg/mL to 496.02 µg/mL in the ABTS assay. The trend for FRAP activities of the extracts tested, did not markedly differ from their DPPH and ABTS

Table 2: Antioxidant activity, total phenolic and total flavonoid contents of extracts from fourteen culinary herbs and spices.

Plant name	DPPH IC_{50} (µg/mL)	ABTS IC_{50} (µg/mL)	FRAP (TEAC)	TPC (mg GAE/g)	TFC (mg QE/g)
Aframomum polyanthum	15.33±2.36^{a}	29.23±6.72^{a}	0.11±0.02^{a}	74.50±4.72^{a}	5.69±1.38^{a}
Anonidium mannii	165.13±18.22^{b}	216.28±5.37^{b}	0.19±0.02^{b}	69.00±3.40^{a,b}	4.16±0.75^{a,b}
Aframomum alboviolaceum	27.86±1.20^{c,d}	34.77±1.18^{a}	0.18±0.02^{b,c}	52.15±4.71^{c}	4.62±0.41^{a,b}
Adansonia digitata (fruit)	8.15±1.01^{d}	9.16±1.22^{c}	0.75±0.04^{d}	237.68±12.99^{d}	16.14±0.18^{c}
Hibiscus sabdariffa	184.00±9.51^{a}	123.52±4.65^{d}	0.11±0.00^{a}	65.52±1.46^{e}	3.20±0.20^{d}
Ocimum gratissimum	228.70±5.23^{f,g}	271.68±11.97^{e}	0.14±0.02^{a,d}	48.96±1.65^{c}	3.89±0.25^{a,b,e}
Tamarindus indica	484.86±9.51^{g}	496.02±12.86^{f}	0.03±0.00^{e}	41.98±7.09^{c,f}	2.98±0.19^{d,f}
Adansonia digitata(leaf)	79.78±5.53^{h}	30.84±4.54^{a}	0.15±0.00^{c}	50.84±1.63^{c}	3.40±0.21^{b,d}
Cyperus esculentus	11.46±1.55^{i}	10.75±0.76^{c}	0.58±0.01^{f}	119.41±9.08^{g}	11.45±0.32^{g}
Sesamum indicum	13.43±1.11^{j}	14.10±1.24^{g}	0.41±0.02^{g}	68.84±4.06^{e,h}	3.80±0.22^{b}
Cinnamomum zeylanicum	234.37±4.61^{j}	248.86±1.06^{e,h}	0.24±0.01^{h}	59.05±2.77^{c,i}	4.57±0.66^{a,b,d}
Corchorus olitorius	158.15±16.84^{j}	145.94±19.55^{i,d}	0.26±0.00^{i}	80.53±4.60^{j}	8.31±0.11^{h}
Sesamum radiatum	20.56±1.07^{k,l}	44.55±2.93^{j}	0.30±0.00^{e}	173.79±6.12^{k}	11.38±0.14^{g}
Aframomum kayserianum	298.81±10.77^{l,m}	270.49±20.21^{e}	0.14±0.02^{j}	54.49±8.50^{l}	6.26±0.59^{a}
Trolox	9.62±0.28^{n}	2.40±0.22^{k}	1.00±0.00^{k}	nd	nd
Ascorbic acid	4.37±0.09^{o}	1.21±0.48^{i}	2.62±0.07^{l}	nd	nd

Values with different letters are significantly different at $p< 0.05$.

scavenging activities; TEAC values varied from 0.03 to 0.75. The free radical scavenging effect of the extract of *A. digitata* in the DPPH assay (IC_{50} value of 8.15 µg/mL) was higher than that of the standard antioxidant trolox (IC_{50} of 9.62 µg/mL. In the ABTS assay, *A. digitata* (fruit), *C.*

esculentus and *S. indicum* had significant antioxidant activities compared to extracts of other plants species, but this antioxidant potency was far less than that of trolox and ascorbic acid.

Results in Table 2 also indicate the TPC and TFC of the spices and culinary herbs analyzed as milligram of gallic acid equivalent per gram of extract and milligram quercetin equivalent per gram of extract respectively. *A. digitata* (fruit) had the highest phenolic and flavonoid content (237.68 mg GAE/g and 16.14 mg QE/g respectively) among the fourteen extracts evaluated. This was followed by *S. radiatum* and *C. esculentus* with TPC values of 173.79 and 119.41 mg GAE/g and TFC contents of 11.38 and 11.45 mg QE/g respectively. Consistent with this finding, *T. indica* with the lowest antioxidant activity in the DPPH and ABTS assays also had the lowest phenolic content (41.98 mg GAE/g).

Results in Table 3 indicate the Pearson's correlation between the total phenolic and total

Table 3: Coefficient of correlation $r^2$and Pearson's correlation coefficients of antioxidant activity (DPPH, FRAP, ABTS), total polyphenol content (TPC) and total flavonoid (TFC) of extracts from fourteen culinary herbs and spices.

		DPPH	ABTS	FRAP	TPC	TFC
r^2	**DPPH**	**1**	**0,977**	-0,586	-0,515	-0,478
p			0,000000002	0,0678	0,0597	0,0836
r^2	**ABTS**		1	-0,558	-0,479	-0,448
p				0,0382	0,0832	0,108
r^2	**FRAP**			1	**0,812**	**0,831**
p					0,0004	0,0002
r^2	**TPC**				1	**0,936**
p						0,0000009
r^2	**TFC**					1

There is a significant correlation between pairs of variables with $p<0.05$.

flavonoids content and antioxidant activity, a statistically significant relationship was observed between TPC, TFC and FRAP. It has been reported that the antioxidant activity of plant

materials is well correlated with the content of their phenolic compounds (Velioglu et al., 1998). Different extracts from the tested spices and herbs have been previously evaluated for their antioxidant activity and similar results to our findings have been reported. Dichloromethane and hexane extracts from *S. sabdariffa* and *C. zeylanicum* had good DPPH radical scavenging activity (Salehi et al., 2013). The methanolic extracts of *Corchorus olitorius,* contained quantities of phenolic and flavonoids compounds (Barku et al.,2013). Similar results were reported by Oriakhi et al. (2014) from the ethanol extract of the leaves of *O. gratissimum* and by Vishwanath et al. (2012) from the ethanol extract of the seeds of *S. indicum*. For *C. esculentus,* antioxidant activity was previously found in the ethanol extract and essential oil of seeds (Bopitiya and Madhujith, 2013; Jing et al., 2013; Vishwanath et al., 2012). The antioxidant potential of *A. digitata* (fruit), *S. radiatum, C. esculentus* and *S. indicum* is demonstrated in this study. Although it is dangerous to extrapolate from *in vitro* to *in vivo* activities it is possible that increasing intake of these herbs could have a therapeutic value in addressing oxidative stress conditions.

3.3. Antimycobacterial activity

The MIC values and the total activity of extracts from fourteen spices and culinary herbs against three fast growing Mycobacterium species strains are shown in Table 4. In general there were not major differences in the activity of the extracys of the different *Mycobacterium* species. The different extract examined had an average MIC of 0.89 mg/mL against M. fortuitum compared to 0.98 mg.mL against *M. aurum* and 1.12 mg/mL against *M. smegmatis.*

Table 4: Minimal inhibitory concentration (MIC in mg/mL) and total activity (TA in mL/g) of extracts from fourteen culinary herbs and spices against mycobacterial strains.

			Microorganisms					
	Extraction	**ME**	***M. smegmatis***		***M. fortuitum***		***M. aurum***	
Plant name	**yield (%)**	**(mg)**	MIC	TA	MIC	TA	MIC	TA
Aframomum polyanthum	3.23	32.3	-	-	1.25	25.84	-	-
Anonidium mannii	3.39	33.9	0.62	54.68	0.31	109.35	1.25	27.12
Aframomum alboviolaceum	6.45	64.5	2.5	25.80	-	-	2.5	25.80
Adansonia digitata (fruit)	3.40	34	1.25	27.20	1.25	27.20	0.31	109.68
Hibiscus sabdariffa	4.94	49.4	1.25	39.52	2.5	19.76	1.25	39.52
Ocimum gratissimum	4.75	47.5	0.31	153.23	0.62	76.61	0.62	76.61
Tamarindus indica	17.98	179.8	0.31	580.00	0.15	1198.67	0.078	2305.13
Adansonia digitata (leaf)	12.17	121.7	1.25	97.36	1.25	97.36	1.25	97.36
Cyperus esculentus	15.21	152.1	-	-	-	-	1.25	121.68
Sesamum indicum	4.87	48.7	0.62	78.55	0.62	78.55	0.62	78.55
Cinnamomum zeylanicum	5.65	56.5	1.25	45.20	0.31	182.26	1.25	45.20
Corchorus olitorius	4.81	48.1	1.25	38.48	-	-	1.25	38.48
Sesamum radiatum	6.67	66.7	2.5	26.68	-	-	-	-
Aframomum kayserianum	4.95	49.5	0.31	159.68	0.62	79.84	0.15	330.00
Rifampicin (µg/mL)			50	nd	3.12	nd	12.5	nd

-: >2.5 mg/mL, nd: not determined

Some of thefourteen extracts were active against the tested organisms with MIC values ranging from 0.078 to >2.5 mg/mL. *T. indica* had potent antimycobacterial activity against *M. aurum* with an MIC value 0.078 mg/mL. Four extracts (*A. alboviolaceum*, *C. esculentus*, *C. olitorius*, S. *radiatum*) had no activity against *M. fortuitum* at the highest concentration tested (MIC >2.5

mg/mL) whilst for the other extracts, MIC values ranged from 0.15 to 2.5 mg/mL. Taking into account the cut-off of the antimicrobial activity of plant extracts of 0.1mg/mL (Eloff, 2004; Kuete and Efferth, 2010), the antimycobacterial activity of the culinary herbs and spices obtained in this study varied from significant to inactive. Furthermore *T. indica* had a total activity of 2305 mL/g (Eloff, 2000), indicating that an extract derived from 1 g of *T. indica* fruits can be diluted to approximately 2.3 L of solvent and still inhibit the growth of *M. aurum*. This result is interesting considering the traditional use of this plant. The antimycobacterial activity of edible plants and spices against *M. tuberculosis* has been previously reported (Tekwu et al., 2012; Sergio et al., 2013), but to best of our knowledge, this is the first report on the antimycobacterial activity of culinary herbs and spices against fast growing *Mycobacterium* species. It has been reported that, activity against the fast growing *M. aurum* is highly predictive of activity against *M. tuberculosis*, as the two species have similar drug sensitivity profiles (Chung et al., 1995). Therefore, the significant activity obtained with *T. indica* against *M. aurum* in this study may be of interest for further screening against pathogenic *Mycobacterium* species.

3.4 Cytotoxic activity

Over the past decade a number of *in vitro* methods have been evaluated with the aim of replacing the mouse bioassay for toxicity testing. Cell culture-based toxicity tests are of interest, having the potential to detect more general cytotoxicity endpoints. In the present study, the toxicity of fourteen extracts from culinary herbs and spices was evaluated on Vero monkey cells by the MTT assay. The LC_{50} values ranged between 33.06 and 770.61 µg/mL (Table 5). According to the National Cancer Institute (United States) plant screening program, a crude extract is generally considered to have *in vitro* cytotoxic activity if the LC_{50} is < 20 µg/mL (Boik, 2001).

Table 5: Cytotoxicity of extracts from fourteen culinary herbs and spices on Vero monkey kidney cells and their selectivity index (SI) against mycobacterial strains.

		Selectivity index (LC_{50} /MIC)		
Plant name	**IC_{50} (µg/ml)**	*Ms*	*Mf*	*Ma*
Aframomum polyanthum	73.39±7.42[a]	>0.03	0.06	>0.03
Anonidium mannii	249.10±11.57[b]	0.40	0.80	0.20
Aframomum alboviolaceum	387.00±9.61[c]	0.15	>0.15	0.15
Adansonia digitata (fruit)	204±0.01[d]	0.16	0.16	0.65
Hibiscus sabdariffa	329.66±3.79[e,c]	0.26	0.13	0.26
Ocimum gratissimum	170.61±70.92[f]	0.55	0.27	0.27
Tamarindus indica	121.46±1.55[g,k]	0.39	0.78	**1.55**
Adansonia digitata(leaf)	606.32±50.46[h]	0.49	0.49	0.49
Cyperus esculentus	770.61±70.92[i]	>0.31	>0.31	0.62
Sesamum indicum	140.00±17.47[k]	0.22	0.22	0.22
Cinnamomum zeylanicum	159.57±7.69[k,l]	0.13	0.51	0.13
Corchorus olitorius	424.76±19.05[m]	0.34	>0.17	0.34
Sesamum radiatum	105.59±2.08[n]	0.04	>0.04	>0.04
Aframomum kayserianum	33.06±1.85[o]	0.11	0.05	0.21
Doxorubicin	4.51±0.57[p]	nd	nd	nd

Values with different letters are significantly different at $p< 0.05$; *Ms: Mycobacterium smegmatis, Ma: Mycobacterium aurum, Mf*: *Mycobacterium fortuitum.*

On the basis of this threshold, all the extracts tested in our study can be considered as safe. This result provides a support on the safety of their traditional use. Nevertheless the value of 33 µg/mL obtained for *Aframomum kayserianum* raises concern about its safety. It is possible that

the acetone extract used may extract more toxic compounds than what could become available in the way this spice is used.

The extract of *T. indica* had the highest selectivity index (SI) of 1.55 with *M. aurum*. In general SI (also called Therapeutic Index) is a measure of potential efficacy versus adverse effects. The higher the selectivity index for a crude extract, the more likely it is that the activity is not due to a general metabolic toxin. An SI > 1 for a crude extract increases the likelihood that its toxic and antibacterial compounds are different (Cho-Ngwa et al., 2010). For most of the extract, the SI values were less than 1; due to their poor antimycobacterial activity effect.

Conclusion

In conclusion, the culinary herbs and spices investigated have a certain level of 15-lipoxygenase inhibitory and antioxidant activity. The cytotoxicity activity shows that with the possible exception of the two *Aframomum* species these herbs are generally not toxic to Vero cells, thus substantiating their safety for using in food especially since only low quantities are used as a spice. This study provides a scientific support for some of the traditional uses; hence suggesting that increasing the intake of these herbs may be helpful in preventing or reducing the progress of some lifestyle-related diseases. Although it is dangerous to extrapolate from *in vitro* to *in vivo* results, the diversity of the pharmacological activities of the *Adansonia digitata* observed in this study suggests that extracts of this plant species may be of value for application in human and animal health.

Acknowledgments

The University of Pretoria provided a postdoctoral fellowship to JPD. The Medical Research Council (Eloff SIR UPRET) and the National Research Foundation (Grant 77228) in South Africa (NRF) provided funding to support this study.

References

Abreu, P.M., Martins, E.S., Kayser, O., Bindseil, K.U., Siems, K., Seemann, A., Frevert, J., 1999. Antimicrobial, antitumor and antileishmania screening of medicinal plants from Guinea-Bissau. Phytomedicine. 6, 187-195.

Abreu, P.M., Noronha, R.G., 1997. Volatile constituents of the rhizomes of *Aframomum alboviolaceum* (Ridley) K. Schum. from Guinea-Bissau. Flavour Fragrance Journal. 12, 79-83.

Akinboro, A., Bakare, A.A., 2013. Spermatotoxic, cytotoxic and genotoxic evaluation of aqueous extract of *Ocimum gratissimum* in albino mice. Open Access Journal of Medicinal and Aromatic Plants. 4, 10-14.

Al-Qarawi, A.A., Al-Damegh, M.A., El-Mougy, S.A., 2003. Hepatoprotective influence of *Adansonia digitata* Pulp. Journal of Herbs, Spices and Medicinal Plants. 10, 1-6.

Awobajo, F.O., Omorodion-Osagie, E., Olatunji-Bello, I.I., Adegoke, O.A., Adeleke, T.L., 2009. Acute oral toxicity test and phytochemistry of some West African medicinal plants. Nigerian Quarterly Journal of Hospital Medicine. 19, 53-58.

Ayafor, J.F., Tchuendem, M.H.K., Nyasse, B., Tillequin, F., Anke, H., 1994. Aframodial and other bioactive diterpenoids from *Aframomum* species. Pure and Applied Chemistry. 66, 2327-2330.

Carter, G.W, Young, P.R, Albert, D.H, Bouska, J, Dyer, R, Bel,l R.L, Summers, J.B, Brooks, D.W., 1991. 5-Lipoxygenase inhibitory activity of Zileuton. Journal of Pharmacology and Experimental Therapeutics. 256:929-937.

Bamishaiye E.I., Bamishaiye O.M., 2011. Tiger nut: as a Plant, its derivatives and benefits. African Journal of Food, Agriculture, Nutrition and Development. 11, 5157-5170.

Bankole, M.A., Shittu, L.J.A., Ahmed, T.A., Bankole, M.N., Shittu, R.K., Kpela, T., Ashiru, O.A., 2007. Synergistic antimicrobial activities of phytoestrogens in crude extracts of two sesame species against some common pathogenic microorganisms. African Journal of Traditional, Complementary and Alternative Medicines. 4, 427-433.

Barku, V.Y.A., Opoku-Boahen, Y., Owusu-Ansah, E., Dayie, N.T.K.D., Mensah, F.E., 2013. I*n-vitro* assessment of antioxidant and antimicrobial activities of methanol extracts of six wound healing medicinal plants. Journal of Natural Sciences Research. 3, 74-80.

Becker, B., 1983. The contribution of wild plants to human nutrition in the Ferlo (Northern Senegal). Agroforestery Systems. 1, 257-267.

Beena, G., Shashi, A., Jain, S.K., Verma, A., 2013. Evaluation of analgesic activity of ethanolic extract of *Tamarindus indica* leaves on experimental animal model. International Journal of Pharmaceutical Sciences and Research. 4, 1994-1997.

Benrezzouk, R., Terencio, M.C., Ferrádiz, M.L., San Feliciano, A., Gordaliza, M., Miguel C.J.M, Puente, M.L, Alcaraz, M.J., 1999. Inhibition of human sPLA2 and 5-lipoxygenase activities by two neo-clerodane diterpenoids Life Science. 64:205-211.

Berker, K.I., Guclu, K., Tor, I., Apak, R., 2007. Comparative evaluation of Fe(III) reducing power-based antioxidant capacity assays in the presence of phenanthroline, batho-phenanthroline, tripyridyltriazine (FRAP), and ferricyanide reagents. Talanta. 72, 1157-1165.

Betti, J., 2004. An ethnobotanical study of medicinal plants among the Baka pygmies in the Dja biosphere reserve, Cameroon. African Study Monographs. 25, 1-27.

Biradar, S., Kangralkar, V., Mandavkar, Y., Thakur, M., Chougule, N., 2010. Antiinflammatory, anti-arthritic, analgesic and anticonvulsant activity of *Cyperus* essential oils. International Journal of Pharmacy and Pharmaceutical Sciences. 2, 112-115.

Boik, J., 2001. Natural Compounds in Cancer Therapy. Princeton, MN, USA: Oregon Medical Press.

Bopitiya, D., Madhujith, T., 2013. Antioxidant activity and total phenolic content of sesame (*Sesamum indicum* L.) seed oil. Tropical Agricultural Research. 24, 296-302.

Brand-Williams W., Cuvelier M E, Berset C, 1995. Use of a free radical method to evaluate antioxidant activity. LWT - Food Science and Technology. 28, 25-30.

Bruneton, J., 1999. Pharmacognosy, Phytochemistry, Medicinal Plants. Lavoisier; 2nd edition Paris.

.

Bujnakova, D., Juhas, S., Faix, S., 2013. The anti-translocation and anti-inflammatory effect of cinnamon oil in mice with TNBS induced colitis. Biologia (Bratislava). 68, 1000-1003.

Cho-Ngwa, F., Abongwa, M., Ngemenya, M.N., Nyongbela, K.D., 2010. Selective activity of extracts of *Margaritaria discoidea* and *Homalium africanum* on *Onchocerca ochengi*. BMC Complementary and Alternative Medicine 10, 2010.

Chung G.A., Aktar, Z., Jackson, S., Duncan, K., 1995. High-throughput screen for detecting antimycobacterial agents. Antimicrobial agents and chemotherapy. 39, 2235-2238.

Costantino, L., Albasini, A., Rastelli, G., Benvenuti, S., 1992. Activity of polyphenolic crude extracts as scavengers of superoxide radicals and inhibitors of xanthine oxidase. Planta Medica. 58, 342-344.

Djeussi, D.E., Noumedem, J.A.K., Seukep, J.A., Fankam, A.G., Voukeng, I.K., Tankeo, S.B., Nkuete, A.H.L., Kuete, V., 2013. Antibacterial activities of selected edible plants extracts against multidrug-resistant Gram-negative bacteria. BMC Complementary and Alternative Medicine. 13, 2013.

Eloff, J.N.2004. Quantifying the bioactivity of plant extracts during screening and bioassay-guided fractionation. Phytomedicine 11, 370-371

Eloff, J.N., 2000. On expressing the antibacterial activity of plant extracts - a small first step in applying scientific knowledge to rural primary health care. South African Journal Science. 96, 116-118.

Eloff, J.N., 1998. A sensitive and quick microplate method to determine the minimal inhibitory concentration of plant extracts for bacteria. Planta Medica. 64, 711-713.

Escalona-Arranz, J.C., Peres-Roses, R., Urdaneta-Laffita, I., Camacho-Pozo, M.I., Rodriguez-Amado, J., Licea-Jimenez, I., 2010. Antimicrobial activity of extracts from *Tamarindus indica* L. leaves. Pharmacognosy Magazine. 6, 242-247.

Feussner, I., Wasternack, C., 2002. The lipoxygenase pathway. Annual Review of Plant Biology. 53, 275-297.

Funk, C., 2006. Lipoxygenase pathways as mediators of early inflammatory events in atherosclerosis. Arteriosclerosis, Thrombosis, and Vascular Biology. 26, 1204-1206.

Handoussa, H., Hanafi, R., Eddiasty, I., El-Gendy, M., El-Khatib, A., Linscheid, M., Mahran, L., Ayoub, N., 2013. Anti-inflammatory and cytotoxic activities of dietary phenolics isolated from *Corchorus olitorius* and *Vitis vinifera*. Journal of Functional Foods. 5, 1204-1216.

Hong, J.W., Yang, G., Kim, Y.B., Eom, S.H., Lew, J.H., Kang, H., 2012. Anti-inflammatory activities of cinnamon water extract *in vivo* and *in vitro* LPS-induced models. BMC Complementary and Alternative Medicine 12, 2012).

Ilhan, S., Savaroglu, F., Colak, F., 2007. Antibacterial and antifungal activity of *Corchorus olitorius* L. (Molokhia) extracts. International Journal of Natural and Engineering Sciences. 1, 59-61.

Jing, S., Ouyang, W., Ren, Z.Y., Xiang, H.X., Ma, Z.X., 2013. The *in vitro* and *in vivo* antioxidant properties of *Cyperus esculentus* oil from Xinjiang, China. Journal of the Sciience of Food and Agriculture. 93, 1505-1509.

Kabore, D., Sawadogo-Lingani, H., Diawara, B., Compaore, C.S., Dicko, M.H., Jakobsen, M., 2011. A review of baobab (*Adansonia digitata*) products: effect of processing techniques, medicinal properties and uses. African Journal of Food Science. 5, 833-844.

Katalinic, V., Mozina, S.S., Generalic, I., Skroza, D., Ljubenkov, I., Klancnik, A., 2013. Phenolic profile, antioxidant capacity, and antimicrobial activity of leaf extracts from six *Vitis vinifera* L. varieties. International Journal of Food Properties. 16, 45-60.

Kuete, V., Fankam, A.G., Wiench, B., Efferth, T., 2013. Cytotoxicity and modes of action of the methanol extracts of six Cameroonian medicinal plants against multidrug-resistant tumor cells. Evidence-based Complementary and Alternative Medicine. 2013, ArteD285903.

Kuete, V., Efferth, T., 2010. Cameroonian medicinal plants: pharmacology and derived natural products. Frontiers in pharmacology. 1, 123.

Lin H.H., Chen J.H., Wang C.J., 2011. Chemopreventive properties and molecular mechanisms of the bioactive compounds in *Hibiscus sabdariffa* Linne. Current Medicinal Chemistry.18, 1245-54.

Lin, J.K., Tsai, S.H., Lin-Shiau, S.Y., 2001. Antiinflammatory and antitumor effects of flavonoids and flavanoids. Drugs of the Future. 26:145-157.

McGaw, L.J., Lall, N., Hlokwe, T.M., Michel, A.L., Meyer, J.J.M., Eloff, J.N., 2008. Purified compounds and extracts from *Euclea* species with antimycobacterial activity against *Mycobacterium bovis* and fast-growing mycobacteria. Biological & Pharmaceutical Bulletin. 31, 1429-1433.

Mehta, J.L., Rasouli, N., Sinha, A.K., Molavi, B., 2006. Oxidative stress in diabetes: A mechanistic overview of its effects on atherogenesis and myocardial dysfunction. International Journal of Biochemistry & Cell Biology. 38, 794-803.

Morton, J., 1987. Tamarind, in: Morton, J., Miami, F. (Eds.), Fruits of Warm Climates. Creative Resource Systems, Inc., Winterville, pp. 115-121.

Mosmann, T., 1983. Rapid Colorimetric Assay for Cellular Growth and Survival - Application to proliferation and cytotoxicity assays. Journal of Immunological Methods. 65, 55-63.

Mpiana, P.T., Tshibangu, D.S.T., Shetonde, O.M., Ngbolua, K.N., 2007. *In vitro* antidrepanocytary actvity (anti-sickle cell anemia) of some Congolese plants. Phytomedicine. 14, 192-195.

Muganza, D.M., Fruth, B.I., Lami, J.N., Mesia, G.K., Kambu, O.K., Tona, G.L., Kanyanga, R.C., Cos, P., Maes, L., Apers, S., Pieters, L., 2012. *In vitro* antiprotozoal and cytotoxic activity of 33 ethonopharmacologically selected medicinal plants from Democratic Republic of Congo. Journal of Ethnopharmacology. 141, 301-308.

Mulaudzi, R.B., Ndhlala, A.R., Kulkarni, M.G., Finnie, J.F., Staden, J.v., 2013. Anti-inflammatory and mutagenic evaluation of medicinal plants used by Venda people against venereal and related diseases. Journal of Ethnopharmacology. 146, 173-179.

Munish, K., Anjoo, K., Sisodia, S.S., 2011. Hepatoprotective activity of *Sesamum indicum* Linn. against CCl 4-induced hepatic damage in rats. International Journal of Pharmaceutical and Biological Archives. 2, 710-715.

Neuwinger, H.D., 2000. African Traditional Medicine: A Dictionary of Plant use and Applications with supplement : search system for diseases. Medpharm Scientific, Health & Fitness. California.

Noumi, E.E.M., 2011. Syphilis ailment: prevalence and herbal remedies in Ebolowa subdivision. South region, Cameroon. International Journal of Biomedical and Pharmaceutical Science. 2, 20-28.

Ordonez, A.A.L., Gomez, J.D., Vattuone, M.A., Isla, M.I., 2006. Antioxidant activities of *Sechium edule* (Jacq.) Swartz extracts. Food Chemistry. 97, 452-458.

Oriakhi, K., Oikeh, E.I., Ezeugwu, N., Anoliefo, O., Aguebor, O., Omoregie, E.S., 2014. Comparative antioxidant activities of extracts of *Vernonia amygdalina* and *Ocimum gratissimum* leaves. Journal of Agricultural Science. 6, 13-20.

Pinto, M.D.C., Tejeda, A., Duque, A.L., Macias, P., 2007. Determination of lipoxygenase activity in plant extracts using a modified ferrous oxidation-xylenol orange assay. Journal of Agricultural Food and Chemistry. 55, 5956-5959.

Pooja, B., Leena, S., Sheema, B., Anupma, M., Sunita, D., 2013. Antibacterial, antioxidant, collagenase inhibition, urease inhibition activities and chemical composition of the essential oil and extracts of *Cinnamomum zeylanicum* bark. International Journal of Phytomedicine. 5, 36-45.

Ranasinghe, P., Jayawardana, R., Galappaththy, P., Constantine, G.R., Gunawardana, N.D.V., Katulanda, P., 2012. Efficacy and safety of 'true' cinnamon (*Cinnamomum zeylanicum*) as a pharmaceutical agent in diabetes: a systematic review and meta-analysis. Diabetic Medicine. 29, 1480-1492.

Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., Rice-Evans, C., 1999. Antioxidant activity applying an improved ABTS radical cation decolorization assay. Free Radical Biology and Medicine. 26, 1231-1237.

Rice-Evans, C.A., Miller, N.J., Paganga, G., 1997. Antioxidant properties of phenolic compounds. Trends in Plant Science. 2, 152-159.

Salehi, P., Asghari, B., Esmaeili, M.A., Dehghan, H., Ghazi, I., 2013. α-Glucosidase and α-amylase inhibitory effect and antioxidant activity of ten plant extracts traditionally used in Iran for diabetes. Journal of Medicinal Plants Research. 7, 257-266.

Samie, A., Nefefe, T., Gundidza, M., Mmbengwa, V., Magwa, M., Mtshali, M.S., 2012. Antimicrobial activities and time kill profiles of five essential oils from Southern Africa against selected bacterial and fungal organisms. African Journal of Pharmacy and Pharmacology. 6, 3086-3095.

Saxena, K., Dube, V., Kushwaha, V., Gupta, V., Lakshmi, M., Mishra, S., Gupta, S., Arora, A., Lakshmi, V., Sharma, R.K., Jain, G.K., Murthy, P.K., 2011. Antifilarial efficacy of *Hibiscus sabdariffa* on lymphatic filarial parasite *Brugia malayi*. Medicinal Chemistry Research. 20, 1594-1602.

Sergio, A., Fabiola, C., Guadalupe, N., Blanca, R, León, H, 2013. Evaluation of antimycobacterium activity of the essential oils of cumin (*Cuminum cyminum*), clove (*Eugenia caryophyllata*), cinnamon (*Cinnamomum verum*), laurel (*Laurus nobilis*) and anis (*Pimpinella anisum*) against *Mycobacterium tuberculosis*. Advances in Biological Chemistry. 3, 480-484.

Seukep, J.A., Fankam, A.G., Djeussi, D.E., Voukeng, I.K., Tankeo, S.B., Noumdem, J.A., Kuete, A.H., Kuete, V., 2013. Antibacterial activities of the methanol extracts of seven Cameroonian dietary plants against bacteria expressing MDR phenotypes. Springerplus. 2, 363.

Sharif, M.R., Alizargar, J., Sharif, A., 2013. Evaluation of the wound healing activity of sesame oil extract in rats. World Journal of Medical Sciences. 9, 74-78.

Sommer, C., 2005. Inflammation and Healing, In: Mattson, C.P. (Ed.), Pathophysiology, 7th ed. Lippincott Williams & Wilkins, pp. 387-398.

Tane, P., Tatsimo, S.D., Ayimele, G.A., Connolly, J.D., 2005. Bioactive metabolites from *Aframomum* species. 11th NAPRECA Symposium Book of Proceedings, Antananarivo, Madagascar. 214-223.

Tanko, Y., Yerima, M., Mahdi, M.A., Yaro, A.H., Musa K.Y., Mohammed, A., 2008. Hypoglycemic activity of methanolic stem bark of *Adansonia digitata* extract on blood glucose levels of streptozocin-induced diabetic wistar rats. International Journal of Applied Research in Natural Products. 2, 32-36.

Tekwu, E.M., Askun, T., Kuete, V., Nkengfack, A.E., Nyasse, B., Etoa, F.X., Beng, V.P., 2012. Antibacterial activity of selected Cameroonian dietary spices ethno-medically used against strains of *Mycobacterium tuberculosis*. Journal of Ethnopharmacology. 142, 374-382.

Thomas, J.M.C., Bahuchet, S., Epelboin, A., Furnis, S., 2003. Encyclopédie Des Pygmées Aka: Techniques, Langage Et Société Des Chasseurs Cueilleurs De La Forêt Centrafricaine (Sud-Centrafrique Et Nord- Congo). , Société d'études linguistiques et anthropologiques de France ed. CNRS, France.

Trouillas, P, Calliste, C.A., Allais, D.P., Simon, A., Marfak, A., Delage, C., Duroux, J.L., 2003. Antioxidant, anti-inflammatory and anti-proliferative properties of sixteen water plant extracts used in the Limousin countryside as herbal teas. Food Chemistry. 80: 399-407.

Velioglu, Y.S., Mazza, G., Gao, L., Oomah, B.D., 1998. Antioxidant activity and total phenolics in selected fruits, vegetables, and grain products. Journal of Agricultural Food and Chemistry. 46, 4113-4117.

Vishwanath, H.S., Anilakumar, K.R., Harsha, S.N., Khanum, F., Bawa, A.S., 2012. *In vitro* antioxidant activity of *Sesamum indicum* seeds. Asian Journal of Pharmaceutical and Clinical Research 5, 56-60.

Wickens, G.E., 1979. The uses of the baobab (*Adansonia digitata L.*) in Africa. In: Kunkel, G. (Ed.), Taxonomic Aspects of African Economic Botany. A.E.T.F.A.T, Las Palmas de Gran Canaria, pp. 27-34.

Wijekoon, M.M.J.O., Rajeev, B., Karim, A.A., Fazilah, A., 2013. Chemical composition and antimicrobial activity of essential oil and solvent extracts of torch ginger inflorescence (*Etlingera elatior* Jack.). International Journal of Food Properties. 16, 1200-1210.

Xu, H., Ping, W.Y., 2010. *In vitro* antitumour activity of *Sesamum indicum* Linn flower extracts. Tropical Journal of Pharmaceutical Research. 9, 455-462.

Yekeen, T.A., Akintaro, O.I., Akinboro, A., Azeez, M.A., 2013. Evaluation of cytogenotoxic and nutrient composition of three commonly consumed vegetables in south-western Nigeria. African Journal of Food, Agriculture, Nutrition and Development. 13, 7452-7466.

Zakaria, Z.A., Somchit, M.N., Zaiton, H., Jais, A.M.M., Sulaiman, M.R., Farah, W.O., Nazaratulmawarina, R., Fatimah, C.A., 2006. The *in vitro* antibacterial activity of *Corchorus olitorius* extracts. International Journal of Pharmacology. 2, 213-215.

Zhang, Q., Zhang, J., Shen, J., Silva, A., Dennis, D.A., Barrow, C.J., 2006. A simple 96-well microplate method for estimation of total polyphenol content in seaweeds. Journal of Applied Phycology. 18, 445-450.