AN ETHNOBOTANICAL, ANTIOXIDANT AND ANTI-INFLAMMATORY STUDY OF
MEDICINAL PLANTS USED IN KAKAMEGA COUNTY, WESTERN KENYA

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OF NAIROBI

SCHOOL OF PHARMACY, DEPARTMENT OF PHARMACOLOGY AND
PHARMACOGNOSY

NOVEMBER, 2016
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I declare that this is my original work and has not been presented in any other university for the award of a degree.

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TABLE OF CONTENTS

DECLARATION .......................................................................................................................................... ii
Approval by Supervisors............................................................................................................................... ii
DECLARATION OF ORIGINALITY FORM ............................................................................................ iii
TABLE OF CONTENTS............................................................................................................................. iv
DEDICATION............................................................................................................................................ vii
ACKNOWLEDGEMENTS ....................................................................................................................... viii
LIST OF TABLES ....................................................................................................................................... ix
LIST OF FIGURES ...................................................................................................................................... x
LIST OF ABBREVIATIONS ...................................................................................................................... xi
DEFINITION OF OPERATIONAL TERMS ............................................................................................. xii
ABSTRACT ............................................................................................................................................... xiii
CHAPTER 1 ................................................................................................................................................. 1
  1.1. Introduction ........................................................................................................................................ 1
  1.2. Problem statement .............................................................................................................................. 2
  1.3. Study justification .............................................................................................................................. 3
  1.4. Objectives .......................................................................................................................................... 3
    1.4.1. General Objective ....................................................................................................................... 3
    1.4.2. Specific Objectives ..................................................................................................................... 3
  1.5. LITERATURE REVIEW .................................................................................................................. 4
    1.5.1. Medicinal plants .......................................................................................................................... 4
    1.5.2. Importance of ethnobotanical surveys ..................................................................................... 6
    1.5.3. The medicinal value of the Kakamega forest .............................................................................. 6
    1.5.4. Plants as sources of antioxidants ................................................................................................. 6
    1.5.5. INFLAMMATION .................................................................................................................... 8
    1.5.6. Oxidative damage as a basis of disease ..................................................................................... 10
CHAPTER 2 ............................................................................................................................................... 11
DEDICATION

This work is dedicated to my parents Mr. Arrton Odongo and Mrs. Maureen Odongo and the people of Kakamega County.
ACKNOWLEDGEMENTS

I am thankful to God for his unending grace, mercies and favor during this journey. He remains to be a faithful God.

I would also like to extend my gratitude to my supervisors Dr. Nelly Mungai, Dr. Piggott Mutai and Dr. Esther Karumi for their constant guidance and understanding towards me and for their invaluable contributions. Thank you very much.

I am grateful to my family for the love that they have showered me with, their support, faith and for pushing me to always pursue more in life. I am forever reminded that unity is strength. You truly are God’s gift to me.

I would also like to acknowledge the assistance and contributions of Dr. Faith Okalebo and Dr. Julia Kimondo in carrying out the anti-inflammatory and antioxidant assays. I am truly thankful.

I am thankful to Mr. Samson Ashira and Mr. Isaiah Olumi for tirelessly identifying the participants of this study across Kakamega County. I would also like to extend my heartfelt gratitude to the Traditional medicine practitioners and villagers who willingly gave out their knowledge.

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LIST OF TABLES

Table 1.1: Some African Phytomedicinals available in the world market ................................. 5
Table 1.2: Antioxidant potential of common herbs and spices................................................... 8
Table 3.1: Medicinal plants used in Kakamega County .......................................................... 48
Table 3.2: Yield on extraction of selected medicinal plants used in Kakamega County .............. 23
Table 3.3: Antioxidant activity of the selected medicinal plants............................................... 49
Table 3.4: Effect of the various treatments on the rat paw volumes at different time points ...... 53
Table 3.5: Difference in effect of the various treatments at different time points ..................... 55
Table 3.6: Difference between the various time points compared to baseline readings .......... 55
Table 3.7: Comparison of the effects of various treatments with the vehicle (10 mg/Kg)........... 56
LIST OF FIGURES

Figure 2.1: Location of Kakamega County on map of Kenya ...................................................... 12

Figure 3.1: Distribution of plants species used as medicine in Kakamega County across various families ........................................................................................................................................... 19

Figure 3.2: Percentages of plant parts used as medicinal plants in Kakamega County .............. 20

Figure 3.3: The main routes of administration for the medicinal preparations ....................... 20

Figure 3.4: Graph showing percent oxidation inhibition of Ascorbic acid................................. 50

Figure 3.5: The percent oxidation inhibition of R. vulgaris and P. fischeri against concentration at 250 μg/mL ................................................................................................................................................. 50

Figure 3.6: The percent oxidation inhibition activity of W. ugandensis against concentration at 500 μg/mL ...................................................................................................................................................... 51

Figure 3.7: The percent oxidation inhibition of methanolic plant extracts against concentration at .................................................................

Figure 3.8: Graph showing mean paw volume against time ......................................................... 54
LIST OF ABBREVIATIONS

AGEs: Advanced glycation end-products

AIDS: Acquired Immunodeficiency Syndrome

BHA: Butylatedhydroxyanisole

BHT: Butylatedhydroxytoluene

CAT: Catalase

CVDs: Cardio vascular diseases

DNA: Deoxyribonucleic acid

DPPH: 2, 2-diphenyl-1-picrylhydrazyl (DPPH) free radical

GPx: Glutathione peroxidase

GSH: Glutathione

HIV: Human Immunodeficiency Virus

IC\textsubscript{50}: Median Inhibition Concentration

NSAIDs: Non steroidal anti-inflammatory drugs

ROS: Reactive Oxygen Species

SOD: Superoxide dismutase

STI: Sexually Transmitted Infection

WHO: World Health Organization
DEFINITION OF OPERATIONAL TERMS

Ethnobotany: the systemic study of the relationship between plants and people in the context of their cultural and social significance.

Ethnopharmacology: a multidisciplinary field of inquiry investigating the anthropological rationale and the pharmacological basis of the medicinal use of plants, animals, fungi, micro-organisms, and minerals by human cultures

IC_{50}: the concentration of an inhibitor where the response is reduced by half.

Medicinal plant: any plant which, in one or more of its organs, contains substances that can be used for therapeutic purposes or as precursors for the synthesis of useful drugs

Phytochemicals: chemical compounds that naturally occur in plants that have protective or disease preventive properties
ABSTRACT

Background
For years, medicinal plants have been used worldwide for the treatment of various diseases. Generally, they are considered safer, easily available and more cost effective compared to conventional medicine. To date, medicinal plants still remain a promising source of natural antioxidant and anti-inflammatory agents to address the increasing burden of diseases related to oxidative damage and inflammation.

Objective
The aim of the present work was to screen medicinal plants commonly used in Kakamega county, western Kenya for their antioxidant and anti-inflammatory activity.

Methodology
An ethnobotanical survey was undertaken to establish medicinal plants used to treat various ailments in Kakamega County. Direct interviews were carried out using structured questionnaires. Plant specimens were collected, identified and voucher specimens deposited at the herbarium at the School of Biological Sciences, University of Nairobi. The plants were air dried at room temperature and ground into powder using a hammer mill (Mu haratta mechanical grinder). Extraction was carried out using methanol. Antioxidant activity was then screened using 2, 2-diphenyl-1-picrylhydrazyl (DPPH) assay while carrageenan induced rat paw edema assay was used to screen for anti-inflammatory activity of the selected medicinal plants.

Results
A total of 94 plant species from 41 families were reported to be used as medicinal plants in Kakamega County by 26 respondents. Most of the plants reported were from the Asteraceae (13.8 %) and Fabaceae (11.7 %) families. Majority of the plants were prepared by boiling or as poultices. 24 plant species were for the first time reported to be used in the area and had not been
previously reported in similar studies within and around the area. Common ailments treated were malaria, stomach aches, skin diseases, joint aches, and sexually transmitted infections. Some of the herbal remedies were used alongside conventional medicines for the treatment of the various chronic ailments such as HIV/AIDS.

Seven plants were selected for screening of antioxidant activity based on their ethno medicinal use and an extensive literature review. The methanolic leaf extracts of *Rhus vulgaris* and *Phyllanthus fischeri* displayed relatively high antioxidant activity with IC$_{50}$ values of 163.63 µg/ml and 182.15µg/ml, respectively. The methanolic leaf extracts of *Senna didymobotrya, Justicia betonica, Warburgia ugandensis, Kalanchoe densiflora* and *Solanum dasyphyllum* had weak antioxidant activity with IC$_{50}$ ranging from 1029 µg/ml to 4051µg/ml.

The methanolic leaf extracts of *Rhus vulgaris* and *Phyllanthus fischeri* were screened for their anti-inflammatory activity using indomethacin as a standard. For indomethacin and *Rhus vulgaris* extract, there was a statistically significant difference in the paw size when compared to the vehicle at all the different time points while *Phyllanthus fischeri* displayed mild activity.

**Conclusion and recommendations**

Medicinal plants are widely used in Kakamega County to treat various ailments. The results of this study support the ethnomedicinal uses of *Rhus vulgaris* and *Phyllanthus fischeri*. These two plants are potential sources of antioxidant and anti-inflammatory agents.

It is recommended that the aqueous extracts of the plants should be tested for antioxidant and anti-inflammatory activities especially given that traditional use entails use of aqueous extracts. Phytochemical studies should be undertaken to isolate compounds responsible for the antioxidant and anti-inflammatory activities.
CHAPTER 1

1.1. Introduction

Over the years, plants have been used by humans as medicine to treat a vast number of diseases. The use of medicinal plants cuts across cultural lines as various traditional systems of medicine such as the Traditional Chinese Medicine, Indian traditional medicine and Japanese traditional medicine have been documented (Fabricant and Farnsworth, 2001). In Africa, the use of Egyptian traditional medicine dates from about 2900 B.C. In most African traditional societies, herbal remedies were often prepared as crude extract of medicinal plant organs such as leaves, roots, flowers and barks (Telefo et al., 2011; Cragg and Newman, 2013).

Today, the popularity of traditional medicine has greatly increased across the world in both developed and developing nations. The World Health Organization estimates that about 80% of the population in developing nations use traditional medicines, most of which are plant based remedies as complementary / alternative medicine (WHO, 2005).

Various factors can be attributed to the upsurge in the use of plant based remedies. They may include: economic considerations such as high cost of conventional medicines, perceived lower toxicity and fewer side effects of plant based medicines as these plants have been used before. To add on to the upsurge is the existence of diseases like cancer and Acquired Immuno Deficiency syndrome (AIDS), to which no cure exists and the emergence of new diseases such as Ebola. The increased cases of drug resistance which are being encountered with the use of conventional medicines have favorably contributed to the use of plant based remedies (Bandaranayake ,2006; Abdullahi , 2011; Pan et al., 2014).
Plants have played an important role in drug discovery. For example vincristine and vinblastine which are used for the treatment of cancer are obtained from *Catharanthus roseus*. Quinine an antimalarial is obtained from *Cinchona ledgeriana* while digoxin is obtained from *Digitalis lanata* and is used as a cardiotonic (Fabricant and Farnsworth, 2001).

There are various ways through which plants can be used as sources of drugs. They include: using the whole plant or part of it as a herbal remedy such as *Ginkgo biloba*, isolating bioactive compounds for direct use as therapeutic agents such as morphine. Plants can also provide raw materials for partial synthesis of drugs with higher activity or lower toxicity or they can be used as molecular models to produce new drugs (Fabricant and Farnsworth, 2001).

Despite the immense health benefits realized from use of plants as medicines, several challenges still exist such as insufficient scientific data to support use of some herbal remedies, lack of standardized formulation of herbal remedies and adulteration of herbal materials. According to the WHO, the assessment of the safety and efficacy of herbal remedies still remains a challenge (WHO, 2005; Ekor, 2014).

In spite of these challenges, medicinal plants have a promising future to act as preventive medicine against various diseases and also as complementary medicine alongside conventional treatments so as to increase efficacy or reduce side effects of conventional therapies (Hassan, 2012).

This study focused on establishing medicinal plants used in Kakamega County in Western Kenya and also screen for their antioxidant and anti-inflammatory activity.
1.2. Problem statement

Oxidation is an inevitable naturally occurring metabolic process that leads to the production of free radicals which cause an array of harmful effects such as structural or functional damage to the cell’s enzymes and genetic material and the development of diseases such as cancers and cardiovascular diseases.

Over the last decade there has been an increase in the number people being diagnosed with diseases related to oxidative damage such as cancer and high death rates due to these diseases have been reported. For example in 2012 there were an estimated 14.1 million new cancer cases diagnosed worldwide and this number is expected to increase to 19.3 million by 2025 (World Cancer Research Fund International (WCRF), 2012).

Furthermore, most of the classical anti-inflammatory agents such as glucocorticoids and non-steroidal anti-inflammatory drugs (NSAIDs) are becoming less acceptable due to serious adverse reactions such as gastric intolerance, bone marrow depression and water and salt retention, due to prolonged use of these drugs (Rodrigues and Montenegro, 2011). This increase in the burden of diseases related to oxidative damage coupled with the high cost of medication and the side effects of these therapies necessitates a need for more effective, affordable and safer remedies. Thus there still exists a demand for new antioxidant and anti-inflammatory agents.

1.3. Study justification

In most cultures, folklore is orally passed down from one generation to another and this poses a threat of complete disappearance of information on traditional medicine (Kipkore et al., 2014). An ethnobotanical survey will promote the preservation of this traditional knowledge through proper documentation of plants used and encourage sustainable use of the medicinal plants.
Studies on the effects of synthetic antioxidants on animal models have revealed the toxicity such as liver toxicity and carcinogenesis related with use of synthetic antioxidants like butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT) and propyl gallate in high doses. (Bauer et al., 2001; Gultekin and Doguc, 2013). These create a need for more natural antioxidants and medicinal plants can act as a suitable source.

Inflammation has been linked to the pathogenesis of various chronic diseases such as cardiovascular diseases, type 2 diabetes, cancers, Alzheimer’s disease and non-alcoholic fatty liver disease (NAFLD) (Campbell, 2015). These findings necessitate more research into inflammation and anti-inflammatory agents.

1.4. Objectives

1.4.1. General Objective

The main aim of the study was to carry out an ethnobotanical study of medicinal plants with potential antioxidant and anti-inflammatory activities in Kakamega County, Western Kenya.

1.4.2. Specific Objectives

The specific objectives of the study were to:

1. Carry out an ethnobotanical survey of the medicinal plants used in Kakamega County.
2. Determine the antioxidant activity of selected medicinal plants using DPPH assay.
3. Determine the anti-inflammatory activity of selected medicinal plants using carrageenan induced rat paw edema assay.
1.5. LITERATURE REVIEW

1.5.1. Medicinal plants

The use of medicinal plants is a practice among humans that has been passed down from one generation to another and plays a role in the development of human cultures and various traditional systems of medicine worldwide. According to the WHO, traditional medicine (TM) is defined as, "the sum total of knowledge, skills and practices based on the theories, beliefs and experiences indigenous to different cultures that are used to maintain health, as well as to prevent, diagnose, improve or treat physical and mental illnesses” (WHO, 2013). Based on fossil records, the use of medicinal plants dates back to the middle Paleolithic age 60000 years ago. These plants had a variety of uses such as food seasoning, weapons and medicines (Hassan, 2012).

Medicinal plants can be described as “any plant which, in one or more of its organs, contains substances that can be used for therapeutic purposes or as precursors for the synthesis of useful drugs”. The therapeutically useful phytochemicals obtained from plants include the alkaloids, flavonoids, tannins and phenolic compounds (Sofowora et al., 2013; Choudhury et al., 2015).

In most plants, the quantity and the composition of bioactive compounds present are influenced by genotype, extraction procedure and environmental conditions (Dai and Mumper, 2010; Vinha et al., 2011). Table 1.1 outlines various phytomedicinals obtained from African plants that are available in the world market.

Plants are a major part of most traditional medicine systems and a variety of conventional drugs have been obtained from plants following ethnobotanical leads from traditional remedies. Natural products and their derivatives represent over 50% of all drugs in clinical use worldwide according to Maridass and Britto (2008).
Table 1.1: Some African Phytomedicinals available in the world market

<table>
<thead>
<tr>
<th>Plant species</th>
<th>Pharmacological action</th>
<th>Constituents</th>
<th>Countries</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ancistrocladus abbreviates</td>
<td>Anti-HIV</td>
<td>Michellamine B</td>
<td>Cameroon and Ghana</td>
<td>(Boyd et al., 1994)</td>
</tr>
<tr>
<td>Chrysanthemum cinerariefolium</td>
<td>Insecticides</td>
<td>Pyrethrins</td>
<td>Kenya, Rwanda, Tanzania, South Africa</td>
<td>(Okigbo and Mmeka, 2006)</td>
</tr>
<tr>
<td>Cinchona succirubra</td>
<td>Anti-malarial</td>
<td>Quinine</td>
<td>West Africa</td>
<td>(Reis and Lipp, 1982)</td>
</tr>
<tr>
<td>Corynanthe pachyceras</td>
<td>Male stimulant</td>
<td>Corynanthidine, corynanthine, yohimbine</td>
<td>Ghana</td>
<td>(Okigbo and Mmeka, 2006)</td>
</tr>
<tr>
<td>Prunus africana</td>
<td>Prostate gland hypertrophy</td>
<td>Sterols, triterpenes, docosanol,</td>
<td>Madagascar, Cameroon, Kenya</td>
<td>(Okigbo and Mmeka, 2006)</td>
</tr>
<tr>
<td>Rauwolfia vomitoria</td>
<td>Tranquilizer, antihypertensive</td>
<td>Reserpine, yohimbine</td>
<td>Nigeria, Zaire, Rwanda</td>
<td>(Okigbo and Mmeka, 2006)</td>
</tr>
<tr>
<td>Syzigium aromaticum</td>
<td>Dental remedy</td>
<td>Eugenol, terpenoids</td>
<td>East Africa, Madagascar</td>
<td>(Elujoba et al., 2005)</td>
</tr>
<tr>
<td>Tamarindus indica</td>
<td>Insecticides</td>
<td>Pectins</td>
<td>Egypt</td>
<td>(Gunasena and Hughes, 2000)</td>
</tr>
</tbody>
</table>
1.5.2. Importance of ethnobotanical surveys

Ethnobotanical surveys are of great relevance as they preserve traditional knowledge through proper documentation. Also they have been proven to be an effective approach to natural and synthetic drug discovery to reveal the hidden potential of plants against various illnesses. They often provide valuable insight during the selection of plants (or specific phytochemicals) to be tested in experimental models of various diseases. Examples of drugs discovered based on their ethnomedicinal use include; etoposide, an antitumor agent from *Podophyllum peltatum* which was used by North American Indians as an emetic and vermifuge. Galegine, an antihyperglycemic agent from *Galega officinalis* L. was traditionally used for the treatment of diabetes. Galegine was vital in the synthesis of the antidiabetic drug metformin (Fabricant and Farnsworth, 2001; Telefo *et al.*, 2011).

In Kenya, traditional knowledge is quite untapped as there are 42 ethnic communities each with different practices on traditional medicine but very few ethnobotanical reports exist to document this knowledge (Kigen *et al.*, 2013).

1.5.3. The medicinal value of the Kakamega forest

Kakamega County is home to the Kakamega forest which is Kenya’s only tropical rainforest and has an abundance of biodiversity with numerous plants being used traditionally for their medicinal value. There are more than 380 plant species recorded to be in the forest (KIFCON 1994; Otieno and Analo, 2012). Some notable medicinal plants include; *Mondia whytei* (Mukombero) whose roots are used as an appetizer and aphrodisiac (Shiracko *et al.*, 2014). *Ocimum kilimandscharicum* (Mwonyi) whose leaves are used for Cold, flu and muscle aches. Based on its traditional use, the Naturub® ointment was developed from purified *Ocimum*
*kilimandscharicum* extracts and is listed in Kenya by the Pharmacy and Poisons Board of Kenya (James, 2010).

### 1.5.4. Plants as sources of antioxidants

Antioxidants are natural or synthetic substances that prevent or delay oxidation. Antioxidants have previously been linked to anti-carcinogenic and anti-aging responses. In plants, antioxidants are often present in low concentrations (Pereira and Federal, 2006).

#### 1.5.4.1. Types of antioxidants

There exist both the endogenous and exogenous antioxidants. Endogenous antioxidants are naturally generated within the body. They include enzymes such as superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and metabolic antioxidants such as glutathione, L-arginine, coenzyme Q10 and transferrin which are products of metabolic processes within the body. Exogenous antioxidants are obtained from food or other supplements as they cannot be produced in the body. Some examples of exogenous antioxidants include products like omega-3 and omega-6 fatty acids, vitamins such as vitamin E and vitamin C (Wilcox *et al.* 2004).

Common synthetic antioxidants such as butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT) and propyl gallate are commonly used as food preservatives to prevent lipid oxidation (Alves *et al.* 2010).

For natural antioxidants, phenolic compounds are mainly responsible for the antioxidant activity. Phenolics can be defined as compounds possessing one or more aromatic rings with one or more hydroxyl groups. They are broadly distributed in the plant kingdom and are the most abundant secondary metabolites of plants, with more than 8,000 phenolic structures currently known,
ranging from simple molecules such as phenolic acids to highly polymerized substances such as tannins (Dai and Mumper, 2010).

1.5.4.2. Plants as sources of antioxidants

Plants have the ability to biosynthesize a wide range of non-enzymatic antioxidants which can counteract the Reactive Oxygen Species (ROS) – induced oxidative damage. Almost two-thirds of the world’s plant species have medicinal value and most of these plants possess favorable antioxidant potential (Krishnaiah et al., 2011). Some of the antioxidants are naturally occurring or are formed in response to abiotic and biotic stress conditions. In response to this condition, plants generate several low molecular weight antioxidants such as vitamin C, vitamin E, phenolic acids and high molecular weight antioxidant secondary metabolites such as tannins that can scavenge free radicals, chelate metal ions and also act as reducing agents. In plants, antioxidant activity is not limited to a particular family or part of a plant (Gupta and Sharma, 2006; Kasote et al., 2015).

Nature, through plants has been a source of dietary antioxidants for years. The discovery and isolation of ascorbic acid, a natural antioxidant created great interest in exogenous antioxidants (Kasote et al., 2015).

Several herbs and spices have shown good antioxidant activity and are well documented. Table 1.2 lists commonly used herbs and spices with their antioxidant properties.
Table 1.2: Antioxidant potential of common herbs and spices

<table>
<thead>
<tr>
<th>Botanical name</th>
<th>Common name</th>
<th>IC50 values</th>
<th>chemical constituents</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Camellia sinensis</em>  (Linn.)</td>
<td>Black tea</td>
<td>$6.7 \pm 0.1\mu g/mL$</td>
<td>(Khalaf <em>et al.</em> 2008).</td>
<td></td>
</tr>
<tr>
<td><em>Zingiber officinale</em></td>
<td>Ginger</td>
<td>$11.3 \pm 0.3\mu g/mL$</td>
<td>(Khalaf <em>et al.</em> 2008).</td>
<td></td>
</tr>
<tr>
<td><em>Piper nigrum</em></td>
<td>Black pepper</td>
<td>$65.1 \pm 1.7\mu g/mL$</td>
<td>(Khalaf <em>et al.</em> 2008).</td>
<td></td>
</tr>
<tr>
<td><em>Curcuma longa</em></td>
<td>Turmeric</td>
<td></td>
<td>Curcumin, beta-sitosterol,camphene</td>
<td>(Gupta and Sharma, 2006)</td>
</tr>
<tr>
<td><em>Glycyrrhiza glabra</em></td>
<td>liquorice</td>
<td></td>
<td>Glycyrrhizin,</td>
<td>(Sam <em>et al.</em> , 2001)</td>
</tr>
<tr>
<td><em>Foeniculum vulgare</em></td>
<td>Fennel</td>
<td></td>
<td>Limonene, estragole, anethole</td>
<td>(Ruberto <em>et al.</em> , 2000)</td>
</tr>
<tr>
<td><em>Allium sativa</em></td>
<td>garlic</td>
<td></td>
<td>flavanoid and steroids</td>
<td>(Narendhirakannan <em>et al.</em> , 2010)</td>
</tr>
<tr>
<td><em>Rosmarinus officinalis</em></td>
<td>Rosemary</td>
<td></td>
<td>1,8-cineole, camphor and α-pinene</td>
<td>(Rašković <em>et al.</em> , 2014)</td>
</tr>
</tbody>
</table>

### 1.5.5. INFLAMMATION

#### 1.5.5.1 Definition and types of inflammation

Adedapo and Ofuegbe (2013) describe inflammation as the response of living tissues to injury which is initiated when a stimulus such as infection, physical or chemical agent produces cellular damage. This damage results in the activation of transcription factors that control the expression of many inflammatory mediators such as eicosanoids, biological oxidants and cytokines.
Inflammation also results in accumulation of plasma fluid and blood cells in sites of injury and cause edema (Igbe et al., 2012; Adedapo and Ofuegbe, 2013).

There are two types of inflammation; acute and chronic inflammation. Acute inflammation is rapid and involves the initial response of the body to harmful stimuli and is characterized by increased movement of plasma and leukocytes from the blood to the injury sites. Chronic inflammation occurs over a long period of time and involves destruction of tissues and increased release of inflammatory mediators (Adedapo and Ofuegbe, 2013).

Despite inflammation being a defense mechanism by the body, the events and mediators involved initiate, maintain or potentiate other diseases. They include: rheumatoid arthritis, asthma, chronic inflammatory bowel diseases, type 2 diabetes, neurodegenerative diseases and cancer. Currently, management of inflammation commonly involves the use of non-steroidal anti-inflammatory drugs, glucocorticoids and immunosuppressant drugs (Sosa et al., 2002; Fürst and Zündorf, 2014).

1.5.5.2. Plants as sources of anti-inflammatory agents

Medicinal plants remain a promising source of anti-inflammatory agents. (Konan et al., 2015). Current anti-inflammatory therapies mostly involve classes of drugs that produce serious side effects such as gastric intolerance, bone marrow depression and water and salt retention, resulting from prolonged use of these drugs (Das et al., 2014). Medicinal plants are believed to be an important source of new chemical substances that are safer and with fewer side effects.

A considerably large number of plants have been scientifically validated to exhibit anti-inflammatory activity. They include; Curcuma longa (tumeric) which contains curcumin that has undergone numerous clinical trials for its anti-inflammatory activity. It’s mechanism of action involves inhibiting pro-inflammatory signaling cascades, such as the NFκB pathways and also
down regulating the secretion of prominent cytokines, like TNFα. Colchicine obtained from *Colchicum autumnale* was approved in 2009 in the United States of America (US) for the treatment of familial Mediterranean fever and prevention of acute gout flares. Capsaicin from *Capsicum* species (Solanaceae) has recently been approved by the authorities in the European Union for use against neuropathic pain in non-diabetic adults and in the US against neuropathic pain associated with postherpetic neuralgia (Fürst and Zündorf, 2014).

*Chamomilla recutita* which is an annual herbaceous plant has been reported to possess good anti-inflammatory, antibacterial and antifungal properties (Vinha *et al.*, 2011). The ethanolic bark extract of *Plumeria rubra* also exhibits anti-inflammatory activity. The various phytochemicals responsible for activity include flavonoids, tannins, alkaloids and terpenoids (Das *et al.*, 2014). Konan *et al.* (2015) reported that mice treated with the extracts of *Gomphrena celosioides* at dose of 100mg/kg showed significant anti-inflammatory activity (Konan *et al.*, 2015).

### 1.5.6. Oxidative damage as a basis of disease

In living organisms, an imbalance between production of free radicals and their destruction results in oxidative stress. Oxidative stress is detrimental to human health as an influx in production of Reactive Oxygen Species (ROS) within the human body leads to cell damage and partial or total functional loss of physiological systems in the body (Gupta and Verma, 2010; Krishnaiah *et al.*, 2011).

Free radicals are implicated in development of various human diseases such as cancer whereby free radicals create chromosomal abnormalities and activate the oncogene thus promoting cancer development (Hossain *et al.*, 2013). In chemical carcinogenesis, the hydroxylation of the DNA bases results in genetic mutations (Valko *et al.*, 2004). Existing evidence supports the role of
oxidative stress in a number of Cardiovascular diseases (CVDs) such as atherosclerosis hypertension, cardiomyopathy and congestive heart failure (Schnabel and Blankenberg, 2007).

For neurological diseases such as Alzheimer’s disease, it is evident that oxidative damage plays a role in loss of neurons and the progression to dementia (Perry and Smith, 2002). Oxidative stress has been implicated in inflammatory lung diseases such as asthma and chronic obstructive pulmonary disease (Repine et al.; 1997). The production of ROS at the inflammation sites leads to joint destruction with increased levels of isopostanes and prostaglandins thus resulting in rheumatoid arthritis (Pham-huy, 2008).
CHAPTER 2

METHODOLOGY
This chapter discusses activities carried out during the ethnobotanical survey and the protocols used in the laboratory.

2.1. ETHNOBOTANICAL FIELD STUDY
An ethnobotanical survey was carried out to establish the medicinal plants used in Kakamega County.

2.1.1. Study design
This study was a qualitative cross-sectional study. It involved data collection on medicinal plants at a specific point in time (Levin, 2006).

2.1.2. Study area
The study was carried out in Kakamega County which is located in the western part of Kenya (Figure 2.1) about 30 km North of the equator. It has a total area of 3,050.3 Km$^2$. This area has a population of 1,660,651 and is largely inhabited by the Luyha community and their major economic activities are farming and fishing. The climate is tropical and high rainfall is often experienced (http://kakamega.go.ke/history/).

The Kakamega forest reserve is also located in Kakamega County, 35 km from Lake Victoria. The forest covers about 230 km$^2$ and currently less than 50 percent is indigenous forest. It has a variety of unique flora and fauna and almost 10 - 20% are unique animal species (Savali, 2000).
There are around 160 tree and shrub species, and over 40 snake species have been identified. The forest is also famous for its diversity of birds: about 367 species have been recorded and they include a mix of lowland and highland species. Some species such as Turner's Eremomela (Eremomela turneri) and Chapins' Flycatcher (Muscicapidae lendu) are threatened. The extensive variety of birds acts as a tourist attraction (Savali, 2000). The forest is very useful to locals as a source of timber, fuel, traditional medicines, food and cultural activities such as circumcision. It is also an important watershed for some of the rivers that flow into Lake Victoria (Kakamega Forest Ecosystem Management Plan 2012-2022).

2.1.3. Study population

The target respondents/ informants were local traditional medicine practitioners (herbalists) and villagers who had practical or empirical knowledge on medicinal plants used as herbal remedies (Telefo et al., 2011).
2.1.4. Recruitment, Inclusion and exclusion criteria

Local leaders well versed with the local environment and indigenous language facilitated contact with the herbalists and villagers who had knowledge on traditional medicine.

The informants were above 18 years and must have lived in Kakamega County for not less than three years. The informants also had to be willing to participate in the study.

Any person who did not meet any of the above criteria was excluded from participating in the study.

2.1.5. Sampling method and sample size

A combination of snowballing and purposive sampling were applied as not everyone sampled randomly would have had the required knowledge. Informants were selected based on their availability and knowledge of medicinal plants (Tongco, 2007). Information was collected until saturation. Saturation was reached when the collection of new data did not yield any new information on the medicinal plants used (Mason, 2010).

Based on the principles of qualitative research methodology, Guest et al. recommend a minimum number of twelve participants for this kind of studies (Guest et al., 2006).

2.1.7. Data collection and management

Data were collected in April 2016 using questionnaire-guided interviews as the main tool of data collection (Appendix II). The data sought included information such as the local name of the plant, ailment treated, part used, length of use and method of preparation (Telefo et al., 2011). Data collected were stored in a password protected word document.
2.2. COLLECTION OF PLANT MATERIAL

Plants were identified by the participants in the field and collected by the study investigator. Voucher specimens were prepared and submitted to the herbarium at the School of Biological Sciences, University of Nairobi, where authentication was done and the voucher specimens were deposited.

For pharmacological assays, the plant materials were collected and air dried in a well aerated room at room temperature. Plant parts used were selected based on their ethnomedicinal use. A mechanical grinder was used to grind the dried plant material into powder form that was suitable for extraction purposes.

2.3. MATERIALS AND EQUIPMENT

2.3.1. Equipment

Plant materials were grounded into powder using a hammer mill (Muharatta mechanical grinder). A PB 3002 delta range top loading balance (Mettler Tpeldo AG Greifensee, Switzerland) was used for weighing samples that were more than 1000 mg. An electronic analytical balance (Shimadzu Au W 220D Kyoto, Japan) was used for weighing materials up to 1000 mg. Filtrations were carried out using Whatman filter paper No. 1 (Whatman International Ltd, Madstone England). A rotary vacuum pump (Heidolph Electro GmbH and Co. KG, Kelheim, Germany), was used to dry the extracts. A spectrophotometer (UV-1800 Spectrophotometer, Shimadzu Corporation) was used to measure the absorbance of extracts used in the DPPH assay for antioxidant activity while a modified plethysmograph was used to measure volume changes in the anti-inflammatory assays.
2.3.2. Chemicals, reagents and solvents

Methanol was used for extraction. Reagents were prepared as described in the established procedures and protocols. Carrageenan (Sigma chemical Co. USA, No. C-4014) was used in the anti-inflammatory assay with indomethacin (Dawa Ltd. Kenya) as a standard and sodium chloride (Sigma Aldrich GmbH Seelze, Germany) was used as the negative control. for the antioxidant assay, 2, 2-diphenyl-1-picrylhydrazyl (DPPH) (Sigma Aldrich GmbH Seelze, Germany) was used and ascorbic acid (Sigma Aldrich GmbH Seelze, Germany) was the positive control.

2.4. EXTRACTION

About 100gm of dry powdered plant material was subjected to cold maceration with 500 ml methanol in 1000 ml conical flask and left for about 24 h at room temperature with occasional shaking. Filtration was done using whatmann No: 1 filter paper and the filtrate was concentrated in a vacuum at 65°C using a rotary evaporator and stored at 4°C for further use.

The percentage yield was obtained using the formula:

\[ \frac{W_2 - W_1}{W_0} \times 100. \]

Where \( W_2 \) is the weight of the extract and the container
\( W_1 \) the weight of the container alone
\( W_0 \) the weight of the initial dried sample

(Thirunavukkarasu et al., 2015)
2.5 PREPARATION OF STOCK SOLUTIONS

2.5.1. For anti-oxidant testing

For the plant extracts, the stock solution was prepared by dissolving 50 mg of the dried extract in 50 ml methanol. DPPH stock solution was prepared by dissolving 9.85 mg of the reagent in 250 ml methanol. Ascorbic acid was prepared by dissolving 5 mg in 50 ml methanol.

2.5.2. For anti-inflammatory testing

For the plant extracts, the stock solution was prepared by dissolving 1000 mg of the dried extract in 20 ml distilled water. 1% carrageenan was prepared by dissolving 1 g in 100 ml distilled water. For saline, 0.09 g was dissolved in 10 ml distilled water. For indomethacin, 1 g was dissolved in 40 ml distilled water.

2.6. PHARMACOLOGICAL SCREENING

2.6.1. 1, 1-diphenyl-2-picrylhydrazyl (DPPH) assay for antioxidant activity

Assay for antioxidant activity was carried out as described by Khalaf (2008) and Scio (2009). The working solutions (50, 75, 100, 250, 500 and 1000 μg/ml) of the plant extracts were prepared from the stock solution using a suitable dilution and were added to 1 ml of 0.002% of DPPH prepared in methanol. These solution mixtures were kept in dark for 30 min and optical density was measured at 517 nm. Ascorbic acid was used as a standard in concentrations ranging from 3.125 to 100 μg/ml. The assay was done in triplicates so as to ensure reproducibility (Khalaf et al., 2008).

The antioxidant activity of the samples was expressed as IC₅₀ (inhibitory concentration), which is defined as the concentration (expressed in μg/ml) of sample required to inhibit the formation of
DPPH radicals by 50%. Extract concentration providing 50% inhibition (IC\textsubscript{50}) was calculated from the graph that plotted inhibition percentage against extract concentration (Scio, 2009).

Inhibition of free radical of DPPH in percentage terms (I\%) was calculated in the following way (Kamkar et al., 2014):

\[
I\% = \left(\frac{A_{\text{blank}} - A_{\text{sample}}}{A_{\text{blank}}}\right) \times 100
\]

Where \(A_{\text{blank}}\) is the absorbance of the control reaction (containing all reagents except the sample) and \(A_{\text{sample}}\) indicates the absorbance of the sample.

### 2.6.2. Assay for anti-inflammatory activity

Carrageenan rat paw edema test was carried out as described in literature (Sawadogo \textit{et al.}, 2006; Igbe \textit{et al.}, 2012).

Adult winster rats were randomly divided into four groups. The test groups (A and B) were treated orally with 1000 mg/kg of the extract. The volume of plant extract administered was calculated as;

\[
V(\text{ml}) = \frac{D(\frac{g}{Kg}) \times P(\text{Kg})}{C(\frac{g}{ml})}
\]

Where D is dose of drug / Kg body weight

\[ P \] is body weight of animal

\[ C \] is concentration of stock solution

The reference group (C) was administered with indomethacin (10 mg/kg) orally. The control group (D) received 10 mL/kg of distilled water. The animals were treated 1 h before injection of 0.1 mL of 1% carrageenan into the sub-plantar tissue of the right hind paw (Igbe \textit{et al.}, 2012). Changes in volume was measured using a phlerthysymograph at 0, 1, 2 and 3 hours following
carrageenan administration and increase in the volume of the right hind paws was taken as an indication of edema (Sawadogo et al., 2006).

The percentage inhibition of the inflammation (hind paw edema) was calculated as

\[
\% \text{ inhibition} = \frac{D_o - D_t}{D_o}
\]

Where: \( D_o \) is the average inflammation (hind paw edema) of the control group of rats at a given time.

\( D_t \) is the average inflammation of the drug treated (i.e. extract or reference) rats at the same time (Sawadogo et al., 2006).
2.7. DATA ANALYSIS

2.7.1. Data from interviews

The information obtained from the interviews was summarized in form of a table to show the following parameters; family, scientific name, popular name in the region (vernacular name), ailment treated, part of plant used, preparation method and length of use (Telefo et al., 2011).

2.7.2. Data from pharmacological assays

Data from the antioxidant assay were analyzed using the Microsoft excel software. A graph showing percentage inhibition against concentration was obtained.

For anti-inflammatory assay, comparison between the treatment groups (controls and extract treated groups) was carried out using one way ANOVA. Results were considered significant when P < 0.05 (Igbe et al., 2012).

2.8. Ethical considerations

The study proposal was approved by the Kenyatta National Hospital/University of Nairobi-Ethical Review Committee (Reference number: KNH-ERC/A/179).

Local leaders, who were well versed with the local environment and indigenous language and knew the traditional healers very well, facilitated contact with the participants.

The investigator explained to the participants the details of the study such as the purpose, the procedures, the benefits and risks involved in the study. After the participants had understood and consented to take part in the study, they were requested to sign the attached consent form (Appendix I).

Information was given by the participants on a voluntary basis as no financial compensation was offered. At times, informants were also required to accompany the researcher to the field to
identify plant species. Informants were free to withdraw from the study at their will and were free to refuse to answer any question that they deemed personal, embarrassing or invading (Telefo et al., 2011).

The information provided was treated confidentially and was used solely for research purposes. Breach of confidentiality was minimized through measures such as collecting the minimum necessary subject identifiers, use of name codes instead of participants’ real names and reducing inappropriate disclosures such that information with third parties was shared only on a need to know basis. Participants were explained to the potential benefits and the Intellectual property rights. Any questions that they had were answered by the investigator.
CHAPTER 3

RESULTS AND DISCUSSION

3.1. ETHNOBOTANICAL FIELD STUDY

The study had twenty six participants. Six of them were male and twenty were female. 38 percent of the participants were aged between 35- 50 years, 47 percent were aged between 50- 65 years while 15 percent were 65 years and above. 45% of the participants had completed primary level education. Most of the participants indicated that they had acquired their knowledge on traditional medicine from close relatives. The participants were from Mumias, Lurambi, Butere, Khwisero, Malava, Ikolomani and Shinyalu constituencies. The subtribes encountered included the kisa, marama, tachoni, tsotso, samia and wanga.

A total of 94 plant species were reported to be used as medicinal plants in the County. The medicinal plants were from 41 families. Most of the plants encountered were from the Asteraceae (13.8 %) and Fabaceae (11.7%) families. The number of species reported for each family were as follows: Asteraceae (13), Fabaceae (11), Euphorbiaceae (7), Solanaceae (5), Lamiaceae (5), Malvaceae (4), Acanthaceae (4), Rubiaceae (3), Anacardiaceae (2), Combretaceae (2), Convolvulaceae (2), Verbenaceae (2), Bignoniaceae (2), Myrtaceae (2), Rutaceae (2), Polygonaceae (2), Phyllanthaceae (2), Apocynaceae(1), Apiaceae (1), Amaranthaceae (1), Asparagaceae (1), Boraginaceae (1), Canellaceae (1), Celastraceae (1), Cucurbitaceae(1), Crassulaceae (1), ,Caesalpiniaceae (1), Caricaceae (1), Chenopodiaceae (1), Meliaceae (1), Lauraceae (1), Pittosporaceae (1), Menispermaceae (1), Poaceae (1), Vitaceae (1), Salicaceae (1), Rosaceae (1), Hypericaceae (1), Tiliaceae (1), Melastomataceae (1), Xanthorrhoeaceae (1).
Asteraceae and Fabaceae families are of great ethnobotanical relevance worldwide. The Asteraceae is the largest plant family while the Fabaceae is the third largest terrestrial plant family based on the number of species (Molares and Ladio, 2012). The Fabaceae family consists of various trees and shrubs and has great importance as a food and medicinal resource. For example, *Crotalaria anagyroides* Kunth. was reported to be used as a cow feed. The nutritional value of the Fabaceae has previously been attributed to their ability to fix atmospheric nitrogen useful for protein synthesis (Rahman *et al*., 2014).

Plant parts most commonly used were leaves (48%), stem/bark (19%) and roots (29%) (Figure 3.2). Leaves were preferred as they were perceived to be an easily renewable source.
Majority of the plants were prepared as decoctions by boiling or as poultices with most plants used in combination. For example stem barks of *Mangifera indica* and *Persea americana* were boiled with the roots of *Carica papaya* and taken orally to remedy syphilis. Another preparation methods encountered were burning of medicinal plants to ash. The main routes of administration were oral and topical (Figure 3.3).
Some of the herbal remedies were used alongside conventional medicines for treatment of chronic diseases such as HIV/AIDS. Other than their medicinal value, some of the plants also had nutritional and veterinary value. For example *Corchorus olitorius* was used as a vegetable.

Some plants were named locally based on their characteristics. For example, *Kalanchoe densiflora* is called Okwamatsi in vernacular meaning “the watery plant” due to its succulent nature; *Corchorus olitorius* is called Omurere due to its slippery nature. The local names of some plants could not be established despite them being used as medicinal plants in the area. This may imply that those plants are not indigenous to the Luhya community or did not have any use historically.

Common ailments treated were malaria, stomach aches and other digestive related diseases, skin diseases, back aches, and sexually transmitted infections.

Most of the plants used were similar to those reported in studies conducted within the area and its environs (Otieno and Analo, 2012; Ochwang’I *et al.*, 2014; Shirakho *et al.*, 2016). Otieno and Analo, (2012) recommended more extensive excursions into the Kakamega forest and its environs to reveal and preserve more information on medicinal plant species, particularly through the involvement of a larger number of key informants. Their study had 9 participants (Otieno and Analo, 2012). This study documented, for the first time 25 plant species which had not been reported in the previous ethnobotany studies for the area. They include; *Alternanthera sessilis* L., *Asparagus setaceus* Willd., *Carisa spinarum* L., *Chenopodium opulifolium* DC., *Crotalaria anagyroides* Kunth., *Crotalaria pallida* Aiton. Hort., *Dichrocephala integrifolia* (L.f.) O. Kuntze, *Cyphostemma ukerewense* (Gilg) Desc., *Hibiscus fuscus* Garcke, *Kalanchoe*
densiflora Rolfe, Maytenus arbutifolia var. sidamoensis, Physalis minima L., Physalis peruviana L., Rubia cordifolia L., Rumex abyssinicus Jacq, Sida cordifolia L., Sida tenuicarpa Vollesen, Solanecio cydonifolius (O. Hoffm.) C. Jeffrey, Solanum dasypylum Schumach., Tarenna graveolens (S.Moore) Bremek., Vepris nobilis Delile, Vernonia hymenolepis A.Rich, Vernonia adoensis Sch. Bip. ex Walp. Var., Vernonia auriculifera Hiern., Tristemma maritiana A. Juss. This may be due to the fact that Kakamega is a large area and some areas may not have been covered in previous studies.

The medicinal use of some of the plants was also found to be similar both locally and also with other parts of the world. For example, Ageratun conyozoides and Bridelia micrantha were reported to be used to treat stomach aches and this was consistent with the findings of Okunade, 2012 and Chinaka and Nkeiruka, 2011. Similarly, Alternanthera sessilis and Phyllanthus fischeri were reported useful in the treatment of skin diseases during this study and this findings are also similar to those of Hossain et al., 2014 and Ochwang’I et al., 2014.

In a previous study, it was reported that due to the restricted harvesting of medicinal plants from the Kakamega forest herbalists in the area and its surroundings had adapted cultivation of medicinal plants (Otieno and Analo, 2012). In this study, four herbalists in this study had cultivated Abrus precatorious, Aloe sp., Fuerstia africana, Phyllanthus fischeri and Warburgia ugandensis in their homesteads.
<table>
<thead>
<tr>
<th>BOTANICAL NAME, FAMILY AND VOUCHER NO.</th>
<th>LOCAL NAME</th>
<th>TRADITIONAL USE</th>
<th>PART USED</th>
<th>PREPARATION</th>
<th>ROUTE OF ADMINISTRATION</th>
<th>REPORTED TRADITIONAL USE</th>
<th>REPORTED PHARMACOLOGICAL ACTIVITY</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abrus precatorius L. (Fabaceae) EAO2016/001</td>
<td>olubinu</td>
<td>Inflamed breasts in breast-feeding mothers family planning coughs</td>
<td>WP</td>
<td>burned to ash</td>
<td>oral</td>
<td>Coughs, oral contraceptive, painful swelling, expectorant (Garaniya and Bapodra, 2014)</td>
<td>anti-oxidative (Gul et al., 2013), anti-inflammatory (Arora et al., 2011), antiimplantation, antispermatoegenic, anticancer (Garaniya and Bapodra, 2014)</td>
</tr>
<tr>
<td>Acacia sp (Fabaceae) EAO2016/002</td>
<td>Eshisisia</td>
<td>boils</td>
<td>L</td>
<td>crushed/boiled</td>
<td>Topical</td>
<td>gonorrhea skin disorders (Kokwaro, 1993)</td>
<td>anti-oxidant antimicrobial (Malviya et al., 2011), analgesic and anti-inflammatory (Afsar et al., 2015)</td>
</tr>
<tr>
<td>Acanthus eminens C.B. Clarke (Acanthaceae) EAO2016/003</td>
<td>lirakhalu</td>
<td>High blood pressure Stomach aches</td>
<td>L, Rt</td>
<td>boiled</td>
<td>oral</td>
<td>antimalarial stomachaches and constipation (Kokwaro, 1993; Awan et al., 2014)</td>
<td>None reported</td>
</tr>
</tbody>
</table>

L= Leaves, S B= Stem-bark, WP = Whole plant, Rt= Root
<table>
<thead>
<tr>
<th>BOTANICAL NAME, FAMILY AND VOUCHER NO.</th>
<th>LOCAL NAME</th>
<th>TRADITIONAL USE</th>
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<th>REPORTED TRADITIONAL USE</th>
<th>REPORTED PHARMACOLOGICAL ACTIVITY</th>
</tr>
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<tbody>
<tr>
<td><em>Ageratum conyzoides</em> Linn. (Asteraceae) EAO2016/005</td>
<td>Stops bleeding after injury, stomach aches, bloody diarrhoea</td>
<td>L, Rt</td>
<td>Crushed/boiled</td>
<td>topical</td>
<td>Wounds and burns (Ming, 1999), antidysenteric, Stomach aches, cough, (Okunade, 2002).</td>
<td>Haemostatic effect (Bamidele <em>et al.</em>, 2010), antioxidant (Dores <em>et al.</em>, 2014) antibacterial and antifungal (singh <em>et al.</em>, 2016)</td>
<td></td>
</tr>
</tbody>
</table>

L= Leaves, S B= Stem-bark, WP = Whole plant, Rt= Root
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<th>REPORTED TRADITIONAL USE</th>
<th>REPORTED PHARMACOLOGICAL ACTIVITY</th>
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<tbody>
<tr>
<td>Aloe sp (Xanthorrhoeaceae) EAO2016/007</td>
<td>eshikaha</td>
<td>Malaria, diabetes, nausea, blood cleanser, acne</td>
<td>L</td>
<td>boiled</td>
<td>oral</td>
<td>Appetizer, Malaria, skin diseases, wounds, blood purifier (Yenesew, 2010)</td>
<td>antibacterial antioxidant (Barandozi, 2013), anti-inflammatory wound healing antidiabetic (Radha and Laxmipriya, 2014)</td>
</tr>
<tr>
<td>Alternanthera sessilis L.(Amaranthaceae) EAO2016/008</td>
<td>eye diseases, cuts, wounds, antidote to snake bite; skin diseases</td>
<td>L</td>
<td>eye drops/crushed</td>
<td>topical</td>
<td>skin diseases, severe pain (Hossain et al., 2014)</td>
<td>antimicrobial wound healing (Sunil et al., 2008) anti-diabetic anti-inflammatory hepatoprotective (Walter et al., 2014)</td>
<td></td>
</tr>
<tr>
<td>Asparagus setaceus (Willd.) (Asparagaceae) EAO2016/009</td>
<td>Tooth aches</td>
<td>Rt</td>
<td>Crush add water</td>
<td>oral</td>
<td>Pneumonia, Coughs, bilhazia (Kokwaro, 1993)</td>
<td>None reported</td>
<td></td>
</tr>
</tbody>
</table>

L= Leaves, S B= Stem-bark, WP = Whole plant, Rt= Root
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<th>REPORTED TRADITIONAL USE</th>
<th>REPORTED PHARMACOLOGICAL ACTIVITY</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aspilia pluriseta Schweinf.(Asteraceae) EAO2016/010</td>
<td>shilamalama</td>
<td>Wound healing</td>
<td>L</td>
<td>crushed / boiled</td>
<td>topical/ oral</td>
<td>Skin disease , Eye problems, wounds</td>
<td>antiviral (Cos ,2002)</td>
</tr>
<tr>
<td>Caesalpinia volkensii Harms(Caesalpiniiaceae) EAO2016/012</td>
<td>Omuchera (ajua)</td>
<td>Headaches</td>
<td>L, Rt</td>
<td>boiled</td>
<td>oral</td>
<td>Malaria (Kuria et al., 2001).</td>
<td>Antimalarial (Kuria et al., 2001), Antinociceptive (Ochieng, 2011) anti-inflammatory (Mwangi et al., 2015)</td>
</tr>
<tr>
<td>Cajanus cajan (L.) Millsp.(Fabaceae) EAO2016/013</td>
<td>Ing’oring’ori</td>
<td>Severe flu</td>
<td>L</td>
<td>boiled</td>
<td>oral</td>
<td>Wounds, diarrhoea, diabetes, laxative, dysentery (Wu et al., 2009)</td>
<td>antioxidant (Wu et al., 2009), antibacterial hypercholesterolemia (Luo et al., 2008), Antidiabetic</td>
</tr>
<tr>
<td>BOTANICAL NAME AND VOUCHER NO.</td>
<td>LOCAL NAME</td>
<td>TRADITIONAL USE</td>
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<tr>
<td><strong>Capsicum frutescens</strong> L.(Solanaceae) EAO2016/014</td>
<td>pilepile</td>
<td>joints</td>
<td>F</td>
<td>crushed/boiled</td>
<td>oral</td>
<td>Throat problems, rheumatism, arthritis, muscular pains (Yenesew, 2010)</td>
<td>anti-inflammatory (pal et al., 2011) antibacterial (Koffi-Nevrya et al., 2012) anti-inflammatory (Jolayemi and Ojewole, 2013).</td>
</tr>
<tr>
<td><strong>Carica papaya</strong> L.(Caricaceae) EAO2016/015</td>
<td>lipapai</td>
<td>Gonorrhea, worms, food</td>
<td>Rt, F</td>
<td>boiled</td>
<td>oral</td>
<td>anthelmintic, gonorrhea, Urinary tract infection (Yenesew, 2010; Yogiraj et al., 2014)</td>
<td>anticancer (Nguyen et al., 2012) anti-inflammatory, antidiabetic (Juárez-Rojop et al., 2014) antibacterial (Doughari et al., 2007)</td>
</tr>
<tr>
<td><strong>Carisa spinarum</strong> L.(Apocynaceae) EAO2016/016</td>
<td>eshikata</td>
<td>Eye infections antihelmintic reduces body fat</td>
<td>Rt</td>
<td>Boiled/crushed</td>
<td>Topical/oral</td>
<td>liver disease, microbial disease (Fatima et al., 2013)</td>
<td>Anthelmintic (Harwansh et al., 2010), antimicrobial (Sanwal et al., 2011)</td>
</tr>
</tbody>
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</thead>
<tbody>
<tr>
<td><em>Centella asiatica</em> L.(Apiaceae) EAO2016/017</td>
<td>neruielala</td>
<td>Tooth ache Ear infections</td>
<td>L. Rt</td>
<td>crushed</td>
<td>Apply on tooth/ear drop</td>
<td>jaundice, inflammation, fever, analgesic (Yenesew, 2010)</td>
<td>analgesic anti-inflammatory (Saha et al., 2013) antimicrobial antifungal (Jagtap et al., 2008)</td>
</tr>
<tr>
<td><em>Chenopodium opulifolium</em> DC. (Chenopodiaceae) EAO2016/018</td>
<td>Olukhosha koshe</td>
<td>Diarrhea, Fever in babies</td>
<td>L. Rt</td>
<td>boiled</td>
<td>oral</td>
<td>Wound, malaria (Namukobe et al., 2011)</td>
<td>None reported</td>
</tr>
<tr>
<td><em>Clerodendrum myricoides</em> Hochst. (Lamiaceae) EAO2016/019</td>
<td>eshirangokho</td>
<td>eye infections</td>
<td>L.</td>
<td>boiled</td>
<td>oral</td>
<td>Malaria, chest pains, rheumatism, gonorrhea, wounds (Yenesew, 2010)</td>
<td>Antimicrobial (Njeru et al., 2016)</td>
</tr>
<tr>
<td><em>Combretum apiculatum</em> Sond (Combretaceae) EAO2016/020</td>
<td>muhungula/ chisala</td>
<td>Gonorrhea, Skin diseases, stops bloody urine</td>
<td>Rt, SB</td>
<td>boiled</td>
<td>oral</td>
<td>conjunctivitis, stomach disorders, acne (Lall and Sharma, 2014), antioxidant (Aderogba et al., 2012)</td>
<td>Antibacterial (Lall and Sharma, 2014), Anti-HIV activity (Asres et al., 2005), Hypoglycaemic effect</td>
</tr>
<tr>
<td><em>Combretum molle</em> R.Br. ex G.Don(Combretaceae) EAO2016/021</td>
<td>muhungula</td>
<td>Gonorrhea, Snake bites, Internal stomach bleeding</td>
<td>SB, Rt</td>
<td>boiled</td>
<td>oral</td>
<td>Anthelmintic, antibacterial (Lall and Sharma, 2014), Antifungal (Yenesew, 2010)</td>
<td>None reported</td>
</tr>
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<tr>
<td>Conyza sumatrensis (Retz.) E.H.Walker(Asteraceae) EAO2016/022</td>
<td>nyangweso/ liposhe</td>
<td>Malaria appetizer, tonsils skin pimples throat infections</td>
<td>L</td>
<td>crushed</td>
<td>oral</td>
<td>malaria (Boniface and Pal, 2013)</td>
<td>antimicrobial (Okorosaye-orubite, 2008), antimalarial (Boniface and Pal, 2013)</td>
</tr>
<tr>
<td>Corchorus olitorius L. (Malvaceae) EAO2016/023</td>
<td>omurere</td>
<td>Vegetable Teething in babies</td>
<td>L</td>
<td>Crushed/ boiled</td>
<td>oral</td>
<td>Toothaches, Sore throat, vomiting , strong bones and teeth, laxative (Kokwaro, 1993)</td>
<td>Antioxidant (Keiko et al., 1999), antibacterial (Pal et al.,2006 )</td>
</tr>
<tr>
<td>Cordia africana Lam.(Boraginaceae) EAO2016/024</td>
<td>omukamari</td>
<td>Bone injuries</td>
<td>SB</td>
<td>boiled</td>
<td>Topical</td>
<td>Broken bones, Venereal diseases (Kokwaro, 1993)</td>
<td>Anti-oxidant, anti-inflammatory, antibacterial (Imam et al., 2015)</td>
</tr>
<tr>
<td>Crotalaria anagyroides Kunth.(Fabaceae) EAO2016/025</td>
<td>Musala kwe ingombe</td>
<td>Cow feed</td>
<td>L</td>
<td>Fodder (Sarwatt, and Mkiwa,1988)</td>
<td>oral</td>
<td>None reported</td>
<td>None reported</td>
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<tr>
<td><em>Croton macrostachyus</em> Delile (Euphorbiaceae) EAO2016/027</td>
<td>Asthma, bleeding wounds, purgative, sore throat, tapeworms, stomach aches</td>
<td>SB, L, Rt boiled/crushed Oral/topical</td>
<td>Coughs, purgative, Wound healing, stops bleeding after birth bleeding (Yenesew, 2010) anti-inflammatory (Matu and Staden, 2003) antimicrobial (Obey et al., 2016)</td>
<td></td>
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<tr>
<td><em>Cucumis figarei</em> Naud. (Cucurbitaceae) EAO2016/028</td>
<td>malaria</td>
<td>L boiled oral</td>
<td>Toothache (Burkill, 1985) antiplasmodial (Muregi et al., 2004)</td>
<td></td>
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<tr>
<td><em>Cuscuta australis</em> R. Br. (Convolvulaceae) EAO2016/029</td>
<td>Typhoid, diarrhoea</td>
<td>WP Crushed add water oral</td>
<td>laxative, anthelmintic, sores measles kidney and liver diseases, eye diseases (Kokwaro, 1993) Hepato-protective (Folarin et al., 2014)</td>
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<tbody>
<tr>
<td><strong>Cymbopogon citratus</strong> (DC.)(Poaceae) EAO2016/030</td>
<td>Majani kho bulimo</td>
<td>Flu/colds</td>
<td>L</td>
<td>boiled</td>
<td>Inhal/oral</td>
<td>gastrointestinal disorders, colic treatment, anxiety, aromatherapy (Scio, 2009)</td>
<td>antimalarial (Tchoumbougnang et al., 2005), antibacterial, anti-inflammatory. (Shah et al., 2011)</td>
</tr>
<tr>
<td><strong>Cyphostemma ukerewense</strong> (Gilg) Desc., (Vitaceae) EAO2016/031</td>
<td>libombola</td>
<td>tonsils</td>
<td>L</td>
<td>crushed</td>
<td>oral</td>
<td>Abscess with maggots (Gradé et al., 2009)</td>
<td>None reported</td>
</tr>
<tr>
<td><strong>Dichrocephala integrifolia</strong> (L.f.) O. Kuntze (Asteraceae) EAO2016/032</td>
<td></td>
<td>oral infection</td>
<td>L</td>
<td>crushed add</td>
<td>oral</td>
<td>Dental infection, tooth extraction (Agbor and Naidoo, 2015)</td>
<td>Antimicrobial, antidiarrheal (Rene et al., 2015)</td>
</tr>
<tr>
<td><strong>Dicliptera laxata</strong> C. B. Clarke (Acanthaceae) EAO2016/033</td>
<td>Lunyasi-lwibituti</td>
<td>diarrhea</td>
<td>Rt, L</td>
<td>Chewed/boiled</td>
<td>oral</td>
<td>Rashes and itching (Kothai and Befirdu, 2012)</td>
<td>anti-inflammatory antinoicceptive (Wolde-Mariam et al., 2013), Antimicrobial activity (Kothai and Befirdu, 2012)</td>
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<tr>
<td><em>Dovyalis macrocalyx</em> Warb. (<em>Salicaceae</em>) EAO2016/034</td>
<td>Shina-muterwa/ likunga</td>
<td>boils</td>
<td>L, Rt</td>
<td>crush</td>
<td>oral</td>
<td>headache, constipation, ulcers (Otieno and Analo 2012)</td>
<td>None reported</td>
</tr>
<tr>
<td><em>Dyschoriste radicans</em> (Hochst. ex A.Rich.) (Acanthaceae) EAO2016/035</td>
<td>esimenenenwa</td>
<td>Itchy skin</td>
<td>L</td>
<td>crushed</td>
<td>topical</td>
<td>stomach aches (Chekole <em>et al.</em>, 2015)</td>
<td>None reported</td>
</tr>
<tr>
<td><em>Erythrina abyssinica</em> Lam. ex DC (Fabaceae) EAO2016/037</td>
<td>Omurembe</td>
<td>Chest problems, blood cleanser, Body swellings</td>
<td>SB, Rt</td>
<td>boiled</td>
<td>oral</td>
<td>Mumps, cramps, abdominal distension, fever, coughs, liver inflammation (Kokwaro 1993)</td>
<td>antioxidant (Kipkore <em>et al.</em>, 2014), anti-inflammatory (Nasimolo, 2013)</td>
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<tr>
<td><em>Euphorbia hirta</em> L. (Euphorbiaceae)</td>
<td>okwambere</td>
<td>asthma</td>
<td>L</td>
<td>boiled</td>
<td>oral</td>
<td>gastroenteric disorders, bronchial and respiratory disorders (Okigbo and Mmeka 2006)</td>
<td>antispasmodic, anti-inflammatory, (Okigbo and Mmeka 2006)</td>
</tr>
<tr>
<td>EAO2016/038</td>
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<td>EAO2016/039</td>
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<td></td>
<td>antioxidant and antiinflammatory (Ezeonwumelu <em>et al.</em>, 2012)</td>
</tr>
<tr>
<td><em>Fuerstia africana</em> T.C.E. Fr. (Lamiaceae)</td>
<td>omwonyo/ Kwa matsai</td>
<td>oral infections</td>
<td>L</td>
<td>boiled</td>
<td>oral</td>
<td>Tooth aches, eye (Kipkoreet <em>et al.</em>, 2014)</td>
<td>antimicrobial (Ngeny <em>et al.</em>, 2013)</td>
</tr>
<tr>
<td>EAO2016/040</td>
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</tr>
<tr>
<td><em>Harungana madagascariensis</em> Lam.ex poir (Hypericaceae)</td>
<td>Omusira/ munamasai</td>
<td>Stomach aches itchy genitals Diarrhoea</td>
<td>SB</td>
<td>boiled</td>
<td>oral</td>
<td>Headache, Malaria, Diarrhoea, gonorrhoea (Kokwaro 1993)</td>
<td>antibacterial (Okoli <em>et al.</em>, 2002) analgesic (Njan 2015)</td>
</tr>
<tr>
<td>EAO2016/041</td>
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<tbody>
<tr>
<td><em>Hibiscus fuscus</em> Garcke(^b) (Malvacea) EAO2016/042</td>
<td>oluvu</td>
<td>stroke, chest problems</td>
<td>L, Rt</td>
<td>Crushed add cold water</td>
<td>oral</td>
<td>Muscle Pull (Namukobe et al., 2011)</td>
<td>Antifungal, antibacterial</td>
</tr>
<tr>
<td><em>Hyptis pectinata</em> L. (Lamiaceae) EAO2016/043</td>
<td>Liliviri/ Lifumire likari</td>
<td>Stomach aches</td>
<td>L, Rt</td>
<td>Crushed/ boiled</td>
<td>oral</td>
<td>throat and skin inflammations, pain, and cancer (Falcao et al., 2013)</td>
<td>Antinociceptive anti-inflammatory (Raymundo et al., 2011)</td>
</tr>
<tr>
<td><em>Ipomea cairica</em> L. (Convolvulaceae) EAO2016/044</td>
<td>olunyiri</td>
<td>colds, measles</td>
<td>L</td>
<td>boiled</td>
<td>oral</td>
<td>Rheumatism, inflammations (Ferreira et al., 2006)</td>
<td>Antioxidant, anti-inflammatory activities (Cho et al., 2004), anti-RSV (respiratory syncytial virus) activity (Ma et al., 2002)</td>
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<tbody>
<tr>
<td><em>Kalanchoe densiflora</em> Rolfe (Crassulaceae) EAO2016/047</td>
<td>okwamatsi</td>
<td>general well being</td>
<td>L</td>
<td>boiled</td>
<td>oral</td>
<td>injuries, wounds, anticancer swelling on skin (Kirui <em>et al.</em>, 2014)</td>
<td>Antimicrobial (Kirui <em>et al.</em>, 2014)</td>
</tr>
<tr>
<td><em>Lantana camara</em> L. (Verbenaceae) EAO2016/048</td>
<td>llantana</td>
<td>Nose bleeding breathing problems</td>
<td>L</td>
<td>crushed</td>
<td>topical</td>
<td>Coughs, sore throat, conjunctitis, toothache, colds inhalant, respiratory diseases (Kokwaro, 1993; Scio, 2009)</td>
<td>Antimicrobial (Naz and Bano, 2013)</td>
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<tbody>
<tr>
<td><em>Mangifera indica</em> L(Anacardiaceae) EAO2016/050</td>
<td>liembe</td>
<td>STIs, Food</td>
<td>SB, L, F</td>
<td>boiled</td>
<td>oral</td>
<td>Gastro intestinal tract disorders, diarrhea, sore throat (Hannan et al., 2013)</td>
<td>anti-inflammatory and analgesic (Garrido et al., 2001) antimicrobial (Kabuki et al., 2000)</td>
</tr>
<tr>
<td><em>Markhamia lutea</em> (Benth) K.Schum.(Bignoniaceae,) EAO2016/051</td>
<td>olusiola</td>
<td>malaria, eye problems, STIs</td>
<td>L, SB, Rt</td>
<td>boiled</td>
<td>oral</td>
<td>Malaria, Syphilis, wounds, conjunctivitis (Kokwaro, 1993)</td>
<td>antioxidant (Narendran et al., 2014), antiplasmodial (Lacroix et al., 2009)</td>
</tr>
<tr>
<td><em>Maytenus arbutifolia</em> var. sidamoensis(Celastraceae) EAO2016/052</td>
<td>Omukunga</td>
<td>Wound healing</td>
<td>L</td>
<td>crushed</td>
<td>topical</td>
<td>Livestock feed (Kokwaro, 1993)</td>
<td>antimicrobial (Orabi et al., 2001)</td>
</tr>
<tr>
<td><em>Melia azedarach</em> L. (Meliaceae) EAO2016/053</td>
<td>mwarubaine</td>
<td>Malaria, Blood cleanser, skin diseases, stomach aches, headaches</td>
<td>L, SB, Rt</td>
<td>boiled</td>
<td>Oral/ topical</td>
<td>Stomach aches, measles, malaria, body pains (Yenesew, 2010)</td>
<td>antimalarial (Rakotoarivelo et al., 2015), antioxidant (Orhan et al., 2012)</td>
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<tr>
<td><strong>Microglossa pyrifolia</strong> <em>(Lam.)</em> Kuntze <em>(Asteraceae)</em> EAO2016/054</td>
<td>ingwe</td>
<td>Expel placenta,</td>
<td>Rt, L, SB</td>
<td>chewed Crushed/ boiled</td>
<td>oral/ topical</td>
<td>Headaches, Colds, Malaria, emetic, fever (Kokwaro, 1993)</td>
<td></td>
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<tr>
<td><strong>Ocimum gratissimum</strong> <em>(L.(Lamiaceae))</em> EAO2016/055</td>
<td>omwonyo</td>
<td>Stomach ulcers, Throat infections Bloody diarrhea</td>
<td>L</td>
<td>boiled</td>
<td>oral</td>
<td>Constipation (Matasyoh et al., 2008)</td>
<td>Antimicrobial, Antioxidant (Matasyoh et al., 2008)</td>
</tr>
<tr>
<td><strong>Persea americana</strong> <em>(Mill.(Lauraceae))</em> EAO2016/056</td>
<td>liovacado</td>
<td>STIs Tooth ache</td>
<td>SB, L, S</td>
<td>boiled/ crushed</td>
<td>oral</td>
<td>Hypertension (Lans, 2006)</td>
<td>Analgesic anti-inflammatory(Adeyemi et al., 2002), antimicrobial (Gomez-Flores et al., 2008)</td>
</tr>
<tr>
<td><strong>Phyllanthus fischeri</strong> <em>(Pax(Euphorbiaceae))</em> EAO2016/057</td>
<td>Olukhala</td>
<td>Skin diseases</td>
<td>L, Rt</td>
<td>Crushed/ boiled</td>
<td>Topical</td>
<td>General body illness AIDS management skin cancer (Kokwaro, 1993; Ochwang’I et al.,</td>
<td>None reported</td>
</tr>
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<tr>
<td>Physalis minima L.(Solanaceae) EAO2016/058</td>
<td>Akhalwa katitii</td>
<td>boils</td>
<td>L</td>
<td>Crush add water</td>
<td>oral</td>
<td>inflammations, enlarged spleen, snakebites</td>
<td>anti-bacterial (Patel et al., 2011)</td>
</tr>
<tr>
<td>Physalis peruviana L.(Solanaceae) EAO2016/059</td>
<td>chest problems</td>
<td>F, L</td>
<td>boiled</td>
<td>oral</td>
<td>asthma, malaria and dermatitis (Wu et al., 2006)</td>
<td>antioxidant anti-inflammatory (Wu et al., 2006)</td>
<td></td>
</tr>
<tr>
<td>Pittosporum mannii Hook. F.(Pittosporaceae) EAO2016/060</td>
<td>Mmonyo</td>
<td>Measles, Oral infections, fever,</td>
<td>L, B</td>
<td>boiled</td>
<td>oral</td>
<td>Malaria, Fever, Stomach ache (Nyongbela et al., 2013)</td>
<td>antioxidant (Momeni et al., 2010)</td>
</tr>
<tr>
<td>Plectranthus barbatus Andrews(Lamiaceae) EAO2016/061</td>
<td>Eshirookha</td>
<td>Chest problems, malaria, skin diseases</td>
<td>L, Rt</td>
<td>boiled</td>
<td>oral</td>
<td>Stomach ache, Fever, wounds, sores, respiratory conditions (Lukhoba et al., 2006)</td>
<td>antimicrobial (Verissimo et al., 2014), antioxidant (Falé et al., 2009)</td>
</tr>
</tbody>
</table>

L= Leaves, S B= Stem-bark, WP = Whole plant, Rt= Root
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<tr>
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<th>REPORTED TRADITIONAL USE</th>
<th>REPORTED PHARMACOLOGICAL ACTIVITY</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Psidium guajava</em> L. (Myrtaceae) EAO2016/062</td>
<td>lipera</td>
<td>STIs, nausea, diarrhoea</td>
<td>B, Rt F</td>
<td>boiled</td>
<td>oral</td>
<td>wounds, pain relief, fever (Gutiérrez <em>et al.</em>, 2008)</td>
<td>antimicrobial (Biswa <em>et al.</em>, 2013)</td>
</tr>
<tr>
<td><em>Rosmarinus officinalis</em> L. (Lamiaceae) EAO2016/065</td>
<td>rosemary</td>
<td>flu</td>
<td>L</td>
<td>boiled</td>
<td>oral/ inhalant</td>
<td>Flavouring food, tonic, boils and wounds, expectorant (Genena, 2008)</td>
<td>Antioxidant, antibacterial (Bozin <em>et al.</em>, 2007)</td>
</tr>
</tbody>
</table>

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<th>REPORTED PHARMACOLOGICAL ACTIVITY</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rubia cordifolia L.(Rubiaceae) EAO2016/066</td>
<td>Oluanda nguvo</td>
<td>Coughs, Ringworms oral infections</td>
<td>L, Rt</td>
<td>burned</td>
<td>oral</td>
<td>skin disorders, wound healing</td>
<td>Antibacterial (Basu et al., 2005)</td>
</tr>
<tr>
<td>Rumex abyssinicus Jacq(Polygonaceae) EAO2016/067</td>
<td>amanyasi</td>
<td>Eye infections, General well being</td>
<td>L</td>
<td>Eye drops/boiled</td>
<td>oral</td>
<td>Pneumonia, Coughs, Stomachache, wound healing (Kokwaro, 1993)</td>
<td>Antimicrobial (Getie et al., 2003), anti-inflammatory (Eguale et al., 2011)</td>
</tr>
<tr>
<td>Rumex usambarensis (Eng.ex Damm.) (Polygonaceae) EAO2016/068</td>
<td>likaachi</td>
<td>Urinary tract infections</td>
<td>L, Rt</td>
<td>boiled</td>
<td>Oral</td>
<td>Peptic ulcers, diarrhoea, Vomiting (Boer et al., 2005)</td>
<td>None reported</td>
</tr>
<tr>
<td>Sapium ellipticum Hochst. ex Krauss(Euphorbiaceae) EAO2016/069</td>
<td>Omuthese</td>
<td>asthma</td>
<td>SB, Rt</td>
<td>boiled</td>
<td>oral</td>
<td>Coughs (Kokwaro, 1993)</td>
<td>Antioxidant (Adesegun et al., 2008)</td>
</tr>
<tr>
<td>Senna didmobotrya (Fresen.) Irwin and Barneby(Fabaceae) EAO2016/070</td>
<td>olubino</td>
<td>Stomach aches</td>
<td>L, Rt</td>
<td>Burned/boiled</td>
<td>oral</td>
<td>Malaria, Headaches, back aches (Yenesew, 2010)</td>
<td>Antibacterial (Niguel and Ndiku, 2015)</td>
</tr>
</tbody>
</table>

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<th>REPORTED TRADITIONAL USE</th>
<th>REPORTED PHARMACOLOGICAL ACTIVITY</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Senna occidentalis</em> L. (Fabaceae) EAO2016/071</td>
<td>coughs</td>
<td>Rt</td>
<td>boiled</td>
<td>Oral</td>
<td>Asthma, coughs, inflammation, malaria (Yenesew, 2010)</td>
<td>anti-inflammatory (Sreejith <em>et al.</em>, 2010)</td>
<td></td>
</tr>
<tr>
<td><em>Sesbania sesban</em> (L.) Merr. (Fabaceae) EAO2016/072</td>
<td>olukhure</td>
<td>Genital infections, constipation</td>
<td>L, SB</td>
<td>Crushed add water/ boiled</td>
<td>topical/ oral</td>
<td>purgative, analgesic, inflammation (Usman <em>et al.</em>, 2013)</td>
<td>Antimicrobial (Gomase, 2013)</td>
</tr>
<tr>
<td><em>Sida cordifolia</em> L. (Malvaceae) EAO2016/073</td>
<td>olulundu</td>
<td>Family planning</td>
<td>Rt, L</td>
<td>Crushed and boiled</td>
<td>oral</td>
<td>Stimulate menstruation, abortifacient, diarrhea, lumbago (muscle and joints pain) (Kokwaro, 1993)</td>
<td>antifertility (Pokale <em>et al.</em>, 2012)</td>
</tr>
</tbody>
</table>

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<th>REPORTED TRADITIONAL USE</th>
<th>REPORTED PHARMACOLOGICAL ACTIVITY</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Solanecio cydonifolius</em> (O. Hoffm.) C. Jeffrey (Asteraceae) EAO2016/075</td>
<td>omwikhalu</td>
<td>Joint pains</td>
<td>L</td>
<td>crushed</td>
<td>oral</td>
<td>None reported</td>
<td>None reported</td>
</tr>
<tr>
<td><em>Solanecio mannii</em> (Hook. f) C. Jeffrey (Asteraceae) EAO2016/076</td>
<td>Likaara/ livokho</td>
<td>Skin diseases</td>
<td>L, Rt</td>
<td>Boiled/ crushed and apply</td>
<td>oral</td>
<td>Skin cancer</td>
<td>Antimicrobial (Mbosso et al., 2010), Antiplasmodial and cytotoxic (Uwitonze, 2010)</td>
</tr>
<tr>
<td><em>Solanum dasyphylum</em> Schumach. (Solanaceae) EAO2016/077</td>
<td>Indula</td>
<td>Induce labour, back aches</td>
<td>Rt, L</td>
<td>boiled</td>
<td>oral</td>
<td>Trypanosomiasis, cough (Bekalo et al., 2009)</td>
<td>None reported</td>
</tr>
<tr>
<td><em>Spathodea campanulata</em> Beauv.(Bignoniaceae) EAO2016/079</td>
<td>omutirisya</td>
<td>Tongue infections</td>
<td>L</td>
<td>crushed</td>
<td>oral</td>
<td>malaria, bacterial infections, antimicrobial (Ofori-Kwakye et al.,2009), respiratory diseases(Zaheer et al., 2011)</td>
<td></td>
</tr>
<tr>
<td>BOTANICAL NAME. FAMILY AND VOUCHER NO.</td>
<td>LOCAL NAME</td>
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<tr>
<td><em>spermacoce princeae</em> (K.Schum.) Verdc. (Rubiaceae) EAO2016/080</td>
<td>Kisunda shikuu</td>
<td>boils</td>
<td>L, Rt</td>
<td>boiled</td>
<td>Skin diseases, Hepatic disease, Bacterial infections</td>
<td>(Kokwaro, 1993)</td>
<td>antibacterial</td>
</tr>
<tr>
<td><em>Syzygium guineense</em> DC. (Myrtaceae) EAO2016/082</td>
<td>Omusioma</td>
<td>menstrual pain</td>
<td>SB</td>
<td>boiled</td>
<td>oral</td>
<td>Abdominal pains/stomach aches</td>
<td>antibacterial</td>
</tr>
<tr>
<td><em>Tagetes minuta</em> L. (Asteraceae) EAO2016/083</td>
<td>inzakha</td>
<td>Skin diseases</td>
<td>L</td>
<td>boiled</td>
<td>Steam bathing</td>
<td>Condiment, colds, respiratory tract inflammations</td>
<td>Antimicrobial</td>
</tr>
<tr>
<td><em>Tarenna graveolens</em> (S.Moore) Bremek.(Rubiaceae) EAO2016/084</td>
<td>oluvambo</td>
<td>Aphrodisiac, Erectile dysfunction</td>
<td>SB, Rt</td>
<td>boiled</td>
<td>oral</td>
<td>Sexual impotence and erectile dysfunction</td>
<td>None reported</td>
</tr>
<tr>
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</tr>
<tr>
<td><em>Tithonia diversifolia</em> Hemsl. (Asteraceae) EAO2016/085</td>
<td>Amaua amalulu</td>
<td>Malaria, Tonsils, jiggers</td>
<td>L</td>
<td>boiled</td>
<td>oral/ topical</td>
<td>Malaria (Goffin et al., 2002)</td>
<td>antiplasmodial (Goffin et al., 2002), Antimicrobial (Ongunfolakan et al., 2010)</td>
</tr>
<tr>
<td><em>Toddalia asiatica</em> (L.) Lam. (Rutaceae) EAO2016/086</td>
<td>oluavari</td>
<td>Stomach aches, Regularizes menses, throat infections</td>
<td>Rt, L, S</td>
<td>boiled</td>
<td>oral</td>
<td>stomach problems, malaria, chest pain, sore throat (Orwa et al., 2007)</td>
<td>Antibacterial, (Karunai et al., 2012)</td>
</tr>
<tr>
<td><em>Triumfetta rhomboidea</em> Jacq. (Tiliaceae) EAO2016/087</td>
<td>omkutsa- mutsatsa</td>
<td>reduce blood flow after birth</td>
<td>Rt</td>
<td>crushed</td>
<td>oral</td>
<td>Expel placenta, Stomach ache, diarrhea (Namukobe et al., 2011)</td>
<td>Antimicrobial, antioxidant (Mevy et al., 2006)</td>
</tr>
<tr>
<td><em>Tristemma maritiana</em> A. Juss (Melastomataceae) (EAO2016/088)</td>
<td>Ovushieni</td>
<td>allergies</td>
<td>crushed</td>
<td>oral</td>
<td>None reported</td>
<td>None reported</td>
<td></td>
</tr>
<tr>
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</tr>
<tr>
<td><em>Vepris nobilis</em> Delile(Rutaceae) EAO2016/089</td>
<td>Indari/olutare</td>
<td>Stomach aches, joints</td>
<td>Rt</td>
<td>boiled</td>
<td>oral</td>
<td>Fever, rheumatism, skin disease, coughs, analgesic, (Kokwaro, 1993)</td>
<td>None reported</td>
</tr>
<tr>
<td><em>Vernonia hymenolepis</em> A.Rich.(Asteraceae) EAO2016/090</td>
<td>oluvulosi</td>
<td>stomachaches</td>
<td>Rt</td>
<td>boiled</td>
<td>oral</td>
<td>Purgative, abdominal pains (Kokwaro, 1993), tooth aches (Onzago et al., 2013)</td>
<td>analgesic (Onzago et al., 2013)</td>
</tr>
<tr>
<td><em>Vernonia adoensis</em> Sch. Bip. ex Walp. Var.(Asteraceae) EAO2016/091</td>
<td>imbulusi/ lusutsa</td>
<td>Eye infections</td>
<td>L, SB, Rt</td>
<td>boiled</td>
<td>Oral</td>
<td>Gonorrhea, malaria, heart and kidney problems (Swamy et al., 2013)</td>
<td>antibacterial (Mabhiza et al., 2016)</td>
</tr>
<tr>
<td><em>Vernonia auriculifera</em> Hiern. (Asteraceae) EAO2016/092</td>
<td>lisavakhwa</td>
<td>Snake bites</td>
<td>L</td>
<td>crushed</td>
<td>topical</td>
<td>Blood clotting, wound healing, Measles (Bekele et al., 2015)</td>
<td>antimicrobial (Bekele et al., 2015)</td>
</tr>
</tbody>
</table>

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3.2. YIELD ON EXTRACTION

*Justicia betonica* (whole plant), *Solanum dasyphylum* (leaves), *Senna didymobotrya* (leaves), *Rhus vulgaris* (leaves), *Warburgia ugandensis* (leaves), *Phyllanthus fischeri* (aerial parts) and *Kalanchoe densiflora* (leaves) were subjected to cold maceration with methanol. The yields are presented in Table 3.2

<table>
<thead>
<tr>
<th>PLANT</th>
<th>% w/ w</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Phyllanthus fischeri</em></td>
<td>5.08</td>
</tr>
<tr>
<td><em>Kalanchoedensiflora</em></td>
<td>5.15</td>
</tr>
<tr>
<td><em>Senna didymobotrya</em></td>
<td>7.47</td>
</tr>
<tr>
<td><em>Solanum dasyphylum</em></td>
<td>7.88</td>
</tr>
<tr>
<td><em>Justicia betonica</em></td>
<td>8.03</td>
</tr>
<tr>
<td><em>Warburgia ugandensis</em></td>
<td>11.77</td>
</tr>
<tr>
<td><em>Rhus vulgaris</em></td>
<td>13.76</td>
</tr>
</tbody>
</table>

From these results, *Rhus vulgaris* methanolic leaf extract gave the highest yield while *Phyllanthus fischeri* methanolic leaf extract had the lowest yield. Maceration was preferred so as to reduce any possibility of thermal decomposition of any thermolabile compounds that may be present.
3.3. DPPH ANTIOXIDANT ASSAY

*Justicia betonica* (whole plant), *Solanum dasypylum* (leaves), *Senna didymobotrya* (leaves), *Rhus vulgaris* (leaves), *Warburgia ugandensis* (leaves), *Phyllanthus fischeri* (aerial parts) and *Kalanchoe densiflora* (leaves) were selected for screening of antioxidant activity based on their ethnomedicinal use and an extensive literature review.

Plants selected based on their ethnomedicinal use were: *Kalanchoe densiflora* used to maintain general well being, *Phyllanthus fischeri* is used to treat skin diseases, *Justicia betonica* is used to treat inflammation and *Senna didymobotrya* is used to treat back aches. Plants selected based on literature review were: *Rhus vulgaris* since other plants belonging to this genus such as *Rhus coriara*, have displayed good antioxidant activity. They contain alkaloids, phenols (Gabr *et al.*, 2014). Similarly for *Solanum dasypylum*, other related species such as *Solanum nigrum* and *Solanum torvum* have shown antioxidant activity (Loganayaki, 2010). For *Warburgia ugandensis*, traditional use reported in literature was the treatment of joints (Kokwaro, 1993). Phytochemical studies showed the plant contains flavonoids (Were *et al.*, 2015).

The methanolic leaf extracts of *Rhus vulgaris* and *Phyllanthus fischeri* displayed good antioxidant activity with IC$_{50}$ of 163.63 μg/mL and 182.15 μg/mL respectively. This is the first report on the antioxidant activity of these two plants. The methanolic leaf extracts of *Senna didymobotrya, Justicia betonica, Warburgia ugandensis, Kalanchoe densiflora* and *Solanum dasypylum* had weak antioxidant activity with IC$_{50}$ ranging from 1029 to 4051 μg/ml (Table 3.3).
The antioxidant activity of a plant extract is mainly determined by its phytoconstituents such as phenolic compounds (Rahman et al., 2013). The differences in the concentrations of these compounds in plants may explain the variations in IC$_{50}$ values observed. Graphs showing percentage inhibition against concentration (µg/mL) of the standard and the various plant extracts were obtained (Figures 3.4- 3.7).

<table>
<thead>
<tr>
<th>PLANT</th>
<th>TRADITIONAL USE</th>
<th>IC$_{50}$ µg/mL</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ascorbic acid (standard)</td>
<td></td>
<td>49</td>
</tr>
<tr>
<td><em>Rhus vulgaris</em></td>
<td>Coughs, colds, abdominal pains</td>
<td>163</td>
</tr>
<tr>
<td><em>Phyllanthus fischeri</em></td>
<td>Skin diseases</td>
<td>182</td>
</tr>
<tr>
<td><em>Solanum dasiphyllum</em></td>
<td>Back aches, induce labour</td>
<td>1029</td>
</tr>
<tr>
<td><em>Kalanchoe densiflora</em></td>
<td>General well being</td>
<td>1407</td>
</tr>
<tr>
<td><em>Senna didymobotrya</em></td>
<td>Headaches, back aches</td>
<td>1434</td>
</tr>
<tr>
<td><em>Justicia betonica</em></td>
<td>Inflammation, muscular pains,</td>
<td>1671</td>
</tr>
<tr>
<td><em>Warburgia ugandensis</em></td>
<td>Stomach ache, fever, malaria</td>
<td>4051</td>
</tr>
</tbody>
</table>

Table 3.3: Antioxidant activity of the selected medicinal plants
Figure 3.4: Graph showing percent oxidation inhibition of Ascorbic acid

Figure 3.5: The percent oxidation inhibition of *R. vulgaris* and *P. fischeri* against concentration at 250 μg/mL

Key: RV- *Rhus vulgaris*, PV- *Phyllanthus fischeri*
Figure 3.6: The percent oxidation inhibition activity of *W. ugandensis* against concentration at 500 μg/mL

Key: WU- *Warburgia ugandensis*

Figure 3.7: The percent oxidation inhibition of methanolic plant extracts against concentration at 1000 μg/mL

The presence of reductive phytochemicals in the plant extracts assayed was indicated by their reducing potential and DPPH scavenging activity. The extracts showed a concentration
dependent scavenging activity. *Rhus vulgaris* had the highest activity while *Warburgia ugandensis* had the lowest activity.

For *Rhus vulgaris* no information on its phytochemistry was found. Probably the compounds responsible for activity maybe phenolic compounds as seen in *Rhus coriaria*, where the activity was linked to the presence of phenolic compounds (Gabr *et. al.*, 2014). Despite *Kalanchoe densiflora* displaying weak antioxidant activity, studies on its phytochemical composition show the presence of tannins, flavonoids and cardiac glycosides (Kirui *et al.*, 2014). Flavonoids are known for their antioxidative effects, thus the weak activity observed may be due to a low concentration of flavonoids. Though *Warburgia ugandensis* is commonly used in the treatment of malaria and is widely famed for its antiplasmodial activity (Wube *et al.*, 2005), it had the weakest antioxidant activity. Phytochemical analysis showed the presence of sesquiterpenes and alkaloids (Wube *et al.*, 2005; Maobe *et al.*, 2013).

### 3.4. ANTI-INFLAMMATORY ACTIVITY

Based on their relatively good antioxidant activity, *Rhus vulgaris* and *Phyllanthus fischeri* were selected for screening of their anti-inflammatory activity. Experimental animals were divided into four groups; negative control group, positive control group and *Rhus vulgaris* and *Phyllanthus fischeri* methanolic leaf extract groups.

The experimental data on the changes in rat paw volume were obtained from all the animals in the different groups, recorded and tabulated. Shapiro-Wilk W test showed that the paw volumes were normally distributed. Table 3.4 presents the mean and standard deviation of the increase in paw volume over a period of one and a half hours. The same results are also presented in Figure 3.8.
Table 3.4: Effect of the various treatments on the paw volumes at different time points

<table>
<thead>
<tr>
<th>Treatment</th>
<th>mean and standard deviation of Increase in paw volumes (ml) over time (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
</tr>
<tr>
<td>Saline (10mg/Kg) (n=6)</td>
<td>0.89±0.145</td>
</tr>
<tr>
<td>Indomethacin (25mg/Kg)</td>
<td>0.945±0.08</td>
</tr>
<tr>
<td>(n=6)</td>
<td>2</td>
</tr>
<tr>
<td>Rhus vulgaris (1000 mg/Kg) (n=3)</td>
<td>0.947±122</td>
</tr>
<tr>
<td>Phyllanthus fischeri (1000 mg/Kg) (n=3)</td>
<td>0.88±0.069</td>
</tr>
<tr>
<td>(n=3)</td>
<td>1</td>
</tr>
</tbody>
</table>

For normal saline, the paw size kept on increasing from baseline to a maximum size at 150 min. thereafter, the mean paw size declined slightly. For indomethacin (25 mg/Kg body weight), the paw size decreased continuously up to 90 min and there after increased. Similarly for the *Phyllanthus fischeri* methanolic leaf extract, the paw size decreased but after 30min, the paw size increased slightly.

The most effective agent was *Rhus vulgaris* methanolic leaf extract that caused a decrease for up to 90 minutes and there after the paw size increased slightly. At 1000 mg/kg, the dose seemed to be more efficacious than indomethacin. These results are presented in figure 3.8.
Carageenan induced inflammation usually occurs in two phases. Phase one involves the release of histamine, serotonin and kinins, which cause vasodilation and increased permeability. Phase two inflammation has been linked to release of chemical mediators over time (Barbosa, 2014).

From the graph (Figure 3.8), *Rhus vulgaris* seemed to be effective only in the acute phase as opposed to indomethacin that had an effect in both the acute and delayed phases. Except at baseline (p = 0.762), there was a statistically significant difference across treatment groups at all the different time points. P values less than 0.05 ($P<0.05$) were considered to be statistically significant. These findings are summarized in Table 3.5
There was a statistically significant difference between treatments (p = 0.005). Similarly, the paw size changed with time (p < 0.001). Paired t-test was done to compare whether for each drug, the paw size was different from the baseline value. The results obtained have been summarized in table 3.6

<table>
<thead>
<tr>
<th>Time (min)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.762</td>
</tr>
<tr>
<td>30</td>
<td>0.046</td>
</tr>
<tr>
<td>60</td>
<td>0.053</td>
</tr>
<tr>
<td>90</td>
<td>0.000</td>
</tr>
<tr>
<td>120</td>
<td>0.010</td>
</tr>
<tr>
<td>150</td>
<td>0.000</td>
</tr>
<tr>
<td>180</td>
<td>0.001</td>
</tr>
</tbody>
</table>
### Table 3.6: Difference between the various time points compared to baseline readings

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Time</th>
<th>Difference from baseline (95% CI)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vehicle (10 mg/Kg body weight)</td>
<td>30vs. 0</td>
<td>-0.003 (-0.103, 0.096)</td>
<td>0.947</td>
</tr>
<tr>
<td></td>
<td>60vs. 0</td>
<td>0.037 (-0.063, -0.136)</td>
<td>0.465</td>
</tr>
<tr>
<td></td>
<td>90vs. 0</td>
<td>0.088 (-0.011, 0.188)</td>
<td>0.080</td>
</tr>
<tr>
<td></td>
<td>120vs. 0</td>
<td>0.093 (-0.006, -0.193)</td>
<td>0.065</td>
</tr>
<tr>
<td></td>
<td>150vs. 0</td>
<td>-0.132 (-0.231, -0.032)</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>180vs. 0</td>
<td>0.13 (0.031, 0.229)</td>
<td>0.011</td>
</tr>
<tr>
<td>Indomethacin (25 mg/Kg body weight)</td>
<td>30vs. 0</td>
<td>-0.212 (-0.311, -0.112)</td>
<td>0.000</td>
</tr>
<tr>
<td></td>
<td>60vs. 0</td>
<td>-0.185 (-0.284, -0.086)</td>
<td>0.000</td>
</tr>
<tr>
<td></td>
<td>90vs. 0</td>
<td>-0.25 (-0.357, -0.159)</td>
<td>0.000</td>
</tr>
<tr>
<td></td>
<td>120vs. 0</td>
<td>-0.101 (-0.201, -0.002)</td>
<td>0.045</td>
</tr>
<tr>
<td></td>
<td>150vs. 0</td>
<td>-0.2 (-0.380, -0.100)</td>
<td>0.010</td>
</tr>
<tr>
<td></td>
<td>180vs. 0</td>
<td>-1.155 (-0.254, -0.056)</td>
<td>0.003</td>
</tr>
<tr>
<td><em>Rhus vulgaris</em> (1000 mg/Kg body weight)</td>
<td>30vs. 0</td>
<td>-0.26 (-0.2, -0.120)</td>
<td>0.000</td>
</tr>
<tr>
<td></td>
<td>60vs. 0</td>
<td>-0.2 (-0.340, 0.06)</td>
<td>0.006</td>
</tr>
<tr>
<td></td>
<td>90vs. 0</td>
<td>-0.34 (-0.480, -0.200)</td>
<td>0.000</td>
</tr>
<tr>
<td></td>
<td>120vs. 0</td>
<td>-0.24 (-0.380, -0.100)</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>150vs. 0</td>
<td>-0.24 (-0.380, -0.100)</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>180vs. 0</td>
<td>-0.233 (-0.373, -0.093)</td>
<td>0.001</td>
</tr>
<tr>
<td><em>Phyllanthus fischeri</em> (1000 mg/Kg body weight)</td>
<td>30vs. 0</td>
<td>-0.14 (-0.280, 0.0)</td>
<td>0.051</td>
</tr>
<tr>
<td></td>
<td>60vs. 0</td>
<td>-0.027 (-0.167, 0.114)</td>
<td>0.706</td>
</tr>
<tr>
<td></td>
<td>90vs. 0</td>
<td>-0.02 (-0.160, -0.120)</td>
<td>0.778</td>
</tr>
<tr>
<td></td>
<td>120vs. 0</td>
<td>-0.0133 (-0.154, 0.127)</td>
<td>0.851</td>
</tr>
<tr>
<td></td>
<td>150vs. 0</td>
<td>-0.04 (-0.180, 0.100)</td>
<td>0.572</td>
</tr>
<tr>
<td></td>
<td>180vs. 0</td>
<td>0.046 (-0.094, 0.187)</td>
<td>0.510</td>
</tr>
</tbody>
</table>

The effect of indomethacin (25 mg/Kg), *Rhus vulgaris* (1000 mg/Kg) and *Phyllanthus fischeri* (1000 mg/Kg) was compared with that of the vehicle and the results were summarized in Table 3.7.
### Table 3.7: Comparison of the effects of various treatments with the vehicle (10 mg/Kg)

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Time</th>
<th>Difference in paw size (95% CI)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Indomethacin (25mg/Kg body weight) vs. vehicle</td>
<td>0</td>
<td>0.055 (-0.078, 0.188)</td>
<td>0.415</td>
</tr>
<tr>
<td></td>
<td>30</td>
<td>-0.153 (-0.287, 0.020)</td>
<td>0.025</td>
</tr>
<tr>
<td></td>
<td>60</td>
<td>-0.167 (-0.30, -0.033)</td>
<td>0.015</td>
</tr>
<tr>
<td></td>
<td>90</td>
<td>-0.292 (-0.425, -0.158)</td>
<td>0.000</td>
</tr>
<tr>
<td></td>
<td>120</td>
<td>-0.14 (-0.273, -0.007)</td>
<td>0.040</td>
</tr>
<tr>
<td></td>
<td>150</td>
<td>-0.257 (-0.390, -0.123)</td>
<td>0.000</td>
</tr>
<tr>
<td></td>
<td>180</td>
<td>-0.23 (-0.363, -0.097)</td>
<td>0.001</td>
</tr>
<tr>
<td>Rhus vulgaris (1000 mg/Kg body weight) vs. vehicle</td>
<td>0</td>
<td>0.057 (-0.107, 0.220)</td>
<td>0.049</td>
</tr>
<tr>
<td></td>
<td>30</td>
<td>0.2 (-0.363, 0.037)</td>
<td>0.017</td>
</tr>
<tr>
<td></td>
<td>60</td>
<td>-0.18 (-0.343, -0.0165)</td>
<td>0.031</td>
</tr>
<tr>
<td></td>
<td>90</td>
<td>-0.372 (-0.531, -0.208)</td>
<td>0.000</td>
</tr>
<tr>
<td></td>
<td>120</td>
<td>-0.277 (-0.440, -0.113)</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>150</td>
<td>-0.363 (-0.527, -0.2)</td>
<td>0.000</td>
</tr>
<tr>
<td></td>
<td>180</td>
<td>-0.307 (-0.470, -0.143)</td>
<td>0.000</td>
</tr>
<tr>
<td>Phyllanthus fischeri (1000 mg/Kg body weight) vs. vehicle</td>
<td>0</td>
<td>-0.01 (0.173, 0.153)</td>
<td>0.904</td>
</tr>
<tr>
<td></td>
<td>30</td>
<td>-0.147 (-0.310, 0.017)</td>
<td>0.078</td>
</tr>
<tr>
<td></td>
<td>60</td>
<td>-0.073 (-0.237, 0.090)</td>
<td>0.375</td>
</tr>
<tr>
<td></td>
<td>90</td>
<td>-0.118 (-0.282, 0.045)</td>
<td>0.154</td>
</tr>
<tr>
<td></td>
<td>120</td>
<td>-0.117 (-0.280, 0.047)</td>
<td>0.160</td>
</tr>
<tr>
<td></td>
<td>150</td>
<td>-0.23 (-0.393, -0.067)</td>
<td>0.06</td>
</tr>
<tr>
<td></td>
<td>180</td>
<td>-0.093 (-0.257, -0.070)</td>
<td>0.260</td>
</tr>
</tbody>
</table>

For indomethacin and *Rhus vulgaris* extract, there was a statistically significant difference in the paw size when compared to the vehicle at all the different time points. On the other hand for *Phyllanthus fischeri*, the difference in paw size when compared to the vehicle was not statistically significant at all the different time points. However the difference was negative thus indicating it has a slight anti-inflammatory effect.

*Phyllanthus fischeri* seems to have low anti-inflammatory activity therefore, higher doses are required. On the other hand, *Rhus vulgaris* has more activity, thus lower doses are required. Various phytochemicals such as alkaloids, tannins, flavonoids have previously been linked to anti-inflammatory activity displayed by various plants (Das et al., 2014).
CHAPTER 4

CONCLUSION AND RECOMMENDATIONS

4.1. CONCLUSION

A total of 94 plant species from 41 botanical families mostly from the Asteraceae (13.8 %) and Fabaceae (11.7 %) were reported to be used as medicinal plants in Kakamega County. Majority of the plants were prepared by boiling or as poultices. Common ailments treated were malaria, stomach aches, skin diseases, joint aches, and sexually transmitted infections. Some of the herbal remedies were used alongside conventional medicines.

Indeed ethnobotanical surveys are an effective approach to reveal the hidden medicinal potential of plants against various illnesses. This study reported for the first time 24 plant species that had previously not been reported in similar studies within and around the area.

The methanolic leaf extracts of *Rhus vulgaris* and *Phyllanthus fischeri* had good antioxidant activity while *Senna didymobotrya*, *Justicia betonica*, *Warburgia ugandensis*, *Kalanchoedensiflora* and *Solanum dasyphylum* had weak antioxidant activity. For anti-inflammatory activity *Rhus vulgaris* had good activity. *Phyllanthus fischeri* displayed some mild anti-inflammatory activity.

These results supported the use of these medicinal plants in the treatment of various diseases related to oxidative damage and inflammation such as *Phyllanthus fischeri* and *Rhus vulgaris*.

4.2 RECOMMENDATIONS

The study identified 94 plant species, some of which their medicinal claims are yet to be scientifically validated thus there is a need for more research into their pharmacological activities so as to give scientific credence for the traditional medicinal plants as both complementary and alternative medicines.
The aqueous extracts of these plants be assayed as most traditional remedies are usually prepared using water. This was not possible during this study due lack of a freeze dryer to dry the aqueous plant extracts thus methanolic plant extracts were used.

Further work on the phytochemical analysis should be carried out to determine the specific compounds responsible for antioxidant and anti-inflammatory activity observed in *Rhus vulgaris* and *Phyllanthus fischeri*.

Despite the *in vitro* antioxidant assay for *Rhus vulgaris* and *Phyllanthus fischeri* showing relatively good activity, there is need to undertake *in vivo* experiments as *in vitro* activity is not always replicated in *vivo*. 
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APPENDICES

Appendix I: Volunteer Information and Prior Informed Consent (PIC) Form

UNIVERSITY OF NAIROBI
COLLEGE OF HEALTH SCIENCES
SCHOOL OF PHARMACY
DEPARTMENT OF PHARMACOLOGY AND PHARMACOGNOSY P. O. Box 19676,
NAIROBI, 00202 TEL. 0202 725099

Title of the research study: An evaluation of the antioxidant and anti-inflammatory activities of medicinal plants commonly used in Kakamega county, western Kenya.

Principal investigator: Elizabeth Amwayi Odongo

Study Location: Kakamega County

Purpose of the Study: To identify commonly used medicinal plants in Kakamega county, western Kenya that have potential antioxidant and anti-inflammatory activity.

Dear Participant,

You have been selected to participate in this study. The study seeks to find out which medicinal plants are used in Kakamega County.

To do this, I will use a questionnaire to ask you a number of questions about you, and your use or knowledge of medicinal plants.

Confidentiality: The information you provide is totally confidential and will only be used for research purposes. Your participation is completely voluntary and you can withdraw from the study after having agreed to participate. You are free to refuse to answer any question you deem personal, embarrassing or invading.
**Potential Benefits** of the study: This study may not have a direct benefit to you but it’s going to help document the medicinal plants that promote wellbeing and how we can preserve the environment and increase its resilience while nurturing them. I am in a position to answer questions you have regarding use of traditional medicines.

**Potential Risk** of the Study: There is a minimal risk of breach of confidentiality but the likelihood of this occurring will be significantly reduced by adopting measures such as collecting the minimum necessary subject identifiers, Use of number codes instead of participants’ names and reducing inappropriate disclosures such that information with third parties will be shared on a need to know basis.

**Intellectual property rights:** Information obtained during this study will be used solely for research purposes. For any further work that may arise from this study, you will be consulted on issues such as access and benefit sharing and you may be required to identify the plants.

**Contacts:**

1. **Elizabeth Amwayi Odongo**

   University of Nairobi, School of Pharmacy

   P. O. Box 19676,

   Nairobi, 00202.

2. **Dr. Nelly N. Mungai**

   University of Nairobi, School of Pharmacy

   P. O. Box 19676,

   Nairobi, 00202.

Should you agree to participate in this study, please sign your name below, indicating that you understand the nature of the study, your responsibilities, inconveniences associated with
voluntary participation and that all your questions and concerns have been answered satisfactorily.

For questions related to your rights as a volunteer or any ethical issues in this research work; you may contact;

**The Secretary, Prof. Chindia**
The KNH/UoN Ethics and Research Committee
P.O. Box 19676-00202 Nairobi
Tel 020-2726300 Ext. 44355
Email: uonknh-erc@uonbi.ac.ke

Consent: I have been fully informed about the study, the risks and benefits of it. I had the opportunity to ask questions which were satisfactorily answered. I therefore consent to voluntarily participate in the study.

Name of participant…………………………………………………………………………………………
Signature/ thumb print of participant……………………………………………………………………
Date………………………………………………………………………………………………………………
Name of researcher/research assistant……………………………………………………………………
Signature/Thumbprint………………………… Date …………………………………
Appendix II: Questionnaire

Section A/ Sehemu ya A

1. Name of Interviewer: ______________________ _________________________
2. Sub County ________________

Section B/ Sehemu ya B

CLIENT IDENTIFICATION DETAILS/ MAELEZO YA KUMTAMBUA MHOJIWA

1. Initials of Client/ Herufiza mhojiwa ________________
2. Unique study number/ Nambari ya kificho
3. Age/ Umri  18-35 ☐ 36-50 ☐ 51-64 ☐ 65 and above/ zaidi ☐
4. Sex /jinsia  Male/ kiume ☐ Female/ kike ☐
5. Residence / makazi ________________________________
6. Religion/ dini Christian/ Mkristo ☐ Muslim/ Muislamu ☐ Traditional/ jadi ☐
7. Highest level of formal Education/ kiwango cha juu cha Elimurasmi
   No Formal Education / hakuenda shule ☐ Primary/ shule ya msingi ☐
   Secondary/ shule ya upili ☐ Tertiary and above/ Elimu ya juu ☐
8. Occupation/ Kazi
   ☐ Unemployed/ sina kazi ☐ Self Employed/ kujiajiri ☐ formal employment/ mishahara
   ☐ Student/ Mwanafunzi ☐ Other/ Mengine (Specify)
Section C/ sehemu ya C

MEDICINAL PLANTS USED /DAWA YA MIMEA YANAYOTUMIKA

9. Regarding medicinal plants used for treatment, fill in the table below stating the plant used (local name), reason for use, part used and method of preparation./ kulingana namimea yanayotumika, tafadhal jaza pelo kueleza matumizi)

<table>
<thead>
<tr>
<th>Plant/ Mmea</th>
<th>Ailment treated/ magonjwa inayotibu</th>
<th>Part used/ sehemu inayotumika</th>
<th>Curative /preventive (yapoonya/yazuia)</th>
<th>Method of preparation /Utaratibu wa maandalizi</th>
<th>duration of use/muda wa matumizi</th>
<th>Comments/ maoni</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
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<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

10. For treatment, do you use/mbinu ya matibabu;

- Medicinal plants only/ dawa ya mimea pekee
- In combination with western medicine/ dawa za magharibi pamoja na dawa ya mimea

11. If yes, how do you use them/ jinsi unavyotumia madawa hayo;

1. I use medicinal plants first and if it fails I go to the western medicine/dawa za mimea kwanza kisha yakishindwa natumia dawa za magharibi

2. I use medicinal plants and western medicine together /mimi hutumia dawa za mimea na dawa za magharibi kwa pamoja
3. I use western medicine first and then if it fails I go to medicinal plants/Mimi hutumiadawa za magharibikwanza na kisha yakishindwa mimi hutumia dawa za mimea

4. For the medicinal plants, I use one kind of herb/ mmea mmoja pekee

5. For the medicinal plants, I use a multiple of herbs./mchanganyiko wa mimea

12. Clarify the conditions under which 1, 2 or 3 occurs (is it dependent on seriousness, acute disease, chronic diseases) or for which the medicinal plants are preferred and for which western medicine is preferred./hali ambayo 1, 2 ama 3 hutendeka
Appendix III: KNH/ERC – Letter of Approval

UNIVERSITY OF NAIROBI
COLLEGE OF HEALTH SCIENCES
P O BOX 19676 Code 00202
Telegram: vargali
Tel:254-20-2726390 Ext 44355

Ref: KNH-ERC/A/179

KENYATTA NATIONAL HOSPITAL
P O BOX 20723 Code 02202
Tel: 72300-8
Fax: 725272
Telegram: MEDSUP, Nairobi

Elizabeth Amwayi Odongo
Reg. No.U56/74026/2014
Dept of Pharmacology and Pharmacognosy
School of Pharmacy
College of Health Sciences
University of Nairobi

Dear Elizabeth

REVISED RESEARCH PROPOSAL- AN EVALUATION OF THE ANTIOXIDANT AND ANTI-INFLAMMATORY ACTIVITIES OF MEDICINAL PLANTS COMMONLY USED IN KAKAMEGA COUNTY, WESTERN KENYA (P30/01/2016)

This is to inform you that the KNH-UoN Ethics & Research Committee (KNH-UoN ERC) has reviewed and approved your above proposal. The approval period is from 30th May 2016 – 29th May 2017.

This approval is subject to compliance with the following requirements:

a) Only approved documents (informed consents, study instruments, advertising materials etc) will be used.
b) All changes (amendments, deviations, violations etc) are submitted for review and approval by KNH-UoN ERC before implementation.
c) Death and life threatening problems and serious adverse events (SAEs) or unexpected adverse events whether related or unrelated to the study must be reported to the KNH-UoN ERC within 72 hours of notification.
d) Any changes, anticipated or otherwise that may increase the risks or affect safety or welfare of study participants and others or affect the integrity of the research must be reported to KNH-UoN ERC within 72 hours.
e) Submission of a request for renewal of approval at least 60 days prior to expiry of the approval period. *(Attach a comprehensive progress report to support the renewal)*
f) Clearance for export of biological specimens must be obtained from KNH-UoN ERC for each batch of shipment.
g) Submission of an *executive summary* report within 90 days upon completion of the study.

This information will form part of the database that will be consulted in future when processing related research studies so as to minimize chances of study duplication and/or plagiarism.

For more details consult the KNH-UoN ERC website [http://www.erc.uonbi.ac.ke](http://www.erc.uonbi.ac.ke)

Protect to discover
Yours sincerely,

PROF ML CHINDIA
SECRETARY, KNH-UoN ERC

c.c. The Principal, College of Health Sciences, UoN
The Deputy Director, CS, KNH
The Assistant Director, Health Information, KNH
The Chair, KNH-UoN ERC
The Dean, School of Pharmacy, UoN
The Chair, Dept of Pharmacology and Pharmacognosy, UoN
Supervisors: Dr. Nelly N. Mungai, Dr. Peggoly C. Mutai, Dr. Esther W. Karumi

Protect to discover