Natural Products Chemistry & Research

Getasetegn and Tefera, Nat Prod Chem Res 2016, 4:4
http://dx.doi.org/10.4172/2329-6836.1000220

Review Article Open Access

Biological Activities and Valuable Compounds from Five Medicinal Plants

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Abstract

Currently, there are more than 85,000 plant species that have been documented for medical use globally. This implies, plant derived natural products hold great promise for discovery and development of new pharmaceuticals in diverse human ailments. Among these divers medicinal species *Aframomum corrorima*, *Commiphora myrrha*, *Nigella sativa*, *Ocimum lamiifolium* and *Olea europaea* are one of very well known valuable plants in serving the local people as spice, flavor, fumigants, and sources of traditional and modern medicines. In modern science their medicinal activities have been corroborated by phytochemical and biological research investigations. As the biological studies revealed they have antibacterial, antioxidant, antifungal, antimicrobial and anti-inflammatory properties even though further investigation required on *in vivo* and animal model to ascertain those *in vitro* activities are really valuable in life. The phytochemical investigations have also shown that these plants are holder of diversified useful compound classes like terpene, steroid, flavonoid, glycoside, fatty acid and alkaloids. However, their pharmacology and pharmacokinetics activities have to be further investigated in order to make sure their metabolic effect upon their usage because some compounds have ability to be metabolized during *in vivo* analysis, and to see whether these compounds have synergic or additive effects. Therefore, the purpose of this review paper is to present the compiled and unified scientific studies which have been done on the above well known medicinal plants about their description, medicinal uses, volatile and extracted active compounds, and chemical structures of isolated compounds in justification with scientific researches.

Keywords: *A. corrorima*; *C. myrrha*; *N. sativa*; *O. lamiifolium*; *O. europaea*

Introduction

According to the WHO traditional medicine is a health practices, approaches, knowledge and beliefs incorporating plants, animals and mineral based medicine, spiritual therapies, manual techniques and exercises, applied singularly or in combination to treat, diagnose and prevent illnesses and maintain well being. The therapeutic use of natural products is the early medical practice. To date in the world, there are more than 85,000 plant species that have been documented for medical use. This indicates the plant derived natural products hold great promise for the discovery and development of new pharmaceuticals in a diverse human ailments [1]. This further implies many medicines widely in used today incorporate ingredients from plants. Almost 71% of new drugs that have been approved since 1981 have directly or indirectly been derived from natural products [2]. Hence, traditional medicinal plants have been greatly contributing in the development of modern medicines. The study revealed approximately 80% of the world's population relies on these traditional plant based medicines for primary health care. The remaining 20% of the world's population also depends on plant products for health care [3].

Ethnobotanical studies are one of the reliable guides for humans in utilizing the abundance of herbal resources that nature has to offer. Fabrican and Farnsworth reported on 122 compounds that obtained from 94 species of plants are being used worldwide in medicine [4]. In addition, 80% of these have had ethnomedical uses which are identical or related to the current use of active elements of plant. As a result, the authors have suggested that ethnomedicinal information must be preserved as a valuable source for novel drug discovery. Still there is a requisite for the development of scientific technology to enhance usage of medicinal plant products even though indigenous plants are known and commonly used as a source of medicines and spices in different countries. The following five indigenous plants *Aframomum corrorima*, *Commiphora myrrha*, *Nigella sativa*, *Ocimum lamiifolium*, and *Olea europaea* are among well known indigenous plants being as a source of medicine, spice, flavor and traditional healer. These plants are also holder of diversified compound classes such as flavonoid, terpenoid, steroid and alkaloid.

Therefore, the main aim of this review paper is to present the compile and unified scientific studies which have been done on the above well known five indigenous plants about their description, medicinal uses, volatile and extracted active compounds, and chemical structures of some isolated compounds.

Aframomum corrorima (Braun) Jansen

Aframomum corrorima (Braun) Jansen or Ethiopian cardamom is a renowned spice and medicinal crop of Zingiberaceae family. It is an Indigenous plant to Ethiopia and other countries. It is a close relative to the widely known Indian cardamom (*Elettaria cardamomum* Maton). The species is native to Ethiopia as well as some parts of Southwestern Sudan, Western Uganda, Tanzania and Eritrea [5-7]. *A. corrorima* locally in Ethiopia is known as "*Korerima*". The dried fruits are traded throughout Ethiopia and also into North East Africa, Arabia and India. The word `Aframomum` is composed from Africa and Amomum, the latter derived from the Arabic "*hamma*" which means "hot, warm" indicating the pungency of the seeds. The specific epithet 'corrorima' refers in Latin form to the local name of the species, "*Korerima*" [7,8].

A. corrorima is a perennial aromatic herb growing about 1-2 m high with scaly of large size, bearing flowers either terminally on

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Received May 16, 2016; **Accepted** June 14, 2016; **Published** June 21, 2016

Citation: Getasetegn M, Tefera Y (2016) Biological Activities and Valuable Compounds from Five Medicinal Plants. Nat Prod Chem Res 4: 220. doi:10.4172/2329-6836.1000220

aerial leaf shoots or from the ground level, red-brown underground rhizomes and leafy stems. It flowers usually from January to December and the fruits mature 2 or 3 months later. It is found in humid or moist forests, slightly shaded, wetter and open places in forests at 1350-2000 m altitude. Naturally, in Ethiopia it grows and cultivated in the forests of south and south western parts of the country such as Gamo Gofa, Debub Omo, Kaffa, Ilubabor, Sidamo, Welega and other regions. Even though farmers are currently collecting this spices plant from the forest and sale at the local market, the destruction of the natural forests for various purposes has significantly reduced the diversity of this shade obligate spices [7,9,10].

Medicinal uses

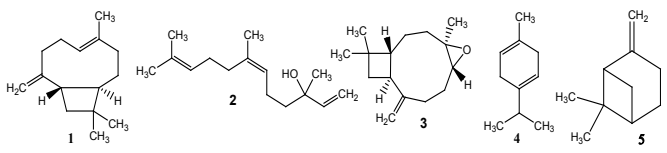
Traditionally "*korerima*" has been used as spices, medicine, an income source and means of soil conservation [11]. It is also used as a carminative, purgative and tonic agent. The seeds are ground and mixed with other spices and used to flavor all kinds of sauces coffee, tea, butter, cottage cheese, bread and "*wot*" [9] and for preparation of "*Berbere*" (hot red pepper) [7].

Ethnobotanical surveys were conducted in the three major "*korerima*" growing regions of southern Ethiopia Gamo Gofa, Debub Omo and Kaffa. Plant parts are used as a medicine for treatment of different aliments. This study revealed 83% of key informants replied that seeds are mostly used as traditional medicine followed by leaves (75%) and rhizomes (72%) [11]. The antioxidant activity of seeds (29.59 μmol/g dry weight (DW)) was significantly higher than that of pods (14.23 μmol/g DW). The total phenolic content in seeds (3.98 mg/g DW) was significantly higher than in pods (1.32 mg/g DW). The phenolic substances including flavonoids and phenolic acids are responsible for the antioxidants activities. The seed of this plant has also antimicrobial and antifungal activities [10,12].

Volatile compounds

Matured green capsules dried on wire mesh bed for 10 days scored maximum oleoresin (10.04% w/w) and essential oil content of dried seeds (5.53%) and essential oil from husks (0.93% v/w) [6]. The composition of essential oil has been investigated by GC and GC/MS. Eighteen components comprising 91% of the oil were identified and majority of them are sesquiterpenes. These are β-caryophyllene (2.11%) (**1**), nerolidol (13.89%) (**2**) and caryophyllene oxide (1.25%) (**3**) [13].

A research was done in Ethiopia to analyze essential oil composition by hydrodistillation. The oil refractive indices were recorded and shown to have 1.494 for leaf and 1.445 for rhizome. The GC and GC/MS analysis of these essential oils identified the presence of 38 (in leaf) and 52 (in rhizome) compounds. The major component of oil from leaf was β-caryophyllene (60.7%) (**1**). The rhizome oil was dominated by γ-terpinene (21.8%) (**4**) and β-pinene (17.6%) (**5**) [11] (Table 1). This essential oil has antiflatulent, virustatic and motility-enhancing effects [7].



Phytochemicals and elements

In the seed part, the levels of essential and non-essential metals were determined using flame atomic absorption spectrometry. As the research revealed the levels of the nutrients were in the following ranges: Ca (1794-2181); Mg (1626-2067); Mn (141-180); Fe (37-46); Zn (12-18); Ni (6.6-8.5); Cu (5.8-8.3); Cr (3.8-5.8) and Co (2.0-2.3) in μg/g. The concentration of non-essential cadmium was in the range from 0.9-1 μg/g while that of lead was below the detection limit of the method [14].

According to Tane et al. some of the Aframomum species are known for the production of labdane diterpenoids, flavonoids, sesquiterpenoids and arylalkaloids which may act against micro-organisms [15]. Solvent extraction yields about 10% oleoresin and this content depends on the solvent used and the raw material. Cardamom oleoresin contains about 52-58% volatile oil and used as flavoring agent [16].

Commiphora myrrha (Nees) Engl.

Myrrh (*Commiphora myrrha* (Nees) Engl.) a yellow fragrant oleo-gum resin is an indigenous plant to Ethiopia and belongs to Burseraceae family. Its resin is collected commercially from South Ethiopia, Somalia, Sudan, Eritrea, Kenya and Arabia. The two species *C. molmol* and *C. myrrha* are synonymous. *C. myrrha* commonly is known in many countries as African myrrha, Somali myrrha, Common myrrha, Herabol myrrha, myrrha [17-19] and in Ethiopia commonly called "*Kerbe*".

C. myrrha is a shrub or tree which growth up to 4 m high. The plant is mainly distributed in Eastern Africa, Arabia and India. It is found in Acacia-Commiphora woodland and bush land on sandy to loamy soil at altitudes 250-1300 m. In Ethiopia it is found in Afar, Bale, Hararge, Sidamo and Somalia. It is also found in north east Kenya and other parts of the world [20].

Myrrh is an aromatic gum resin of the plant stem resinous exudates` of different *Commiphora spp.* that is used as medicinally active extract. Even though it has been collected from various species of the genus *Commiphora* such as *C. africana*, *C. erythraea*, *C. gileadensis*, *C. habessinica*, *C. hodai*, *C. kua*, *C. quadricincta*, *C. schimperi* and *C. truncate*, the best quality has been obtained from the specie of *C. myrrha* [21,22]. Myrrh consists of 7%-17% volatile oil, 25%-40% resin, 57%-61% gum, and 3%-4% impurities [22].

Medicinal uses

Myrrh is used in traditional medicine particularly by women who fumigate their body for health care [22], as spices and flavor [17]. Ethnobotanical survey indicated that it has many medicinal activities and has been used to treat various diseases such as amenorrhea, ache, dysmenorhhea, tumors, fever, stomach complaints, gall bladder, chest ailments, snake and scorpion bites [23]. The British Herbal Compendium allow the use of myrrh tincture as gargle to treat phayngitis and tonsillitis, as mouth wash for gingivitis and ulcers, and externally to treat skin inflammations. In France its topical use is approved for treatment of small wounds, for nasal congestin from the common cold. The German Standard License for myrrh tincture indicates its use for inflammations of the gums and mouth mucosa because it contains large amounts of tannins [21,24].

Other than the ethnobotanical survey, the laboratory based researches have revealed myrrh is used as antibacterial, antiseptic, antitumor, antifungal and anaesthetic activity [25]. Antimicrobial activity of natural extracts and pure compounds can be detected by observing the growth response of various micro-organisms to samples that are placed in contact with them. The research findings suggested that extracts and isolated compounds of myrrh resin could be useful for preventing and treating human gynecologic cancer disease. *In vitro* research shown 85% EtOH and petroleum ether extract significantly

Plant name	Compounds name	Comd. No.	References
Aframomum corrorima	β-Caryophyllene	**1**	[11,13]
	Nerolidol	**2**	
	Caryophyllene oxide	**3**	
	γ-Terpinene	**4**	
	β-Pinene	**5**	
Commiphora myrrha	Germacrene B	**6**	[20,21,28-30,32,34]
	Furanodiene	**7**	
	Lindestrene	**8**	
	Curzerene	**9**	
	β-Elemene	**10**	
	Germacrone	**11**	
	Furanoeudesma-1,3-diene	**12**	
	Curzerenone	**13**	
	Furanodiene-6-one	**14**	
	Neomyrrhaol	**15**	
	Sandaracopimaric acid	**16**	
	Abietic acid	**17**	
	2-Methoxy-5-acetoxyfruranogermacr-1(10)-en-6-one	**18**	
	Dehydroabietic acid	**19**	
	Myrrhterpenoid K	**20**	
	Myrrhterpenoid L	**21**	
	Myrrhterpenoid M	**22**	
	Myrrhterpenoid N	**23**	
	Furanogermacra-1(10)*Z*,4*Z*-dien-6-one	**24**	
	Commiferin	**25**	
	Quercetin	**26**	
	β-Sitosterol	**27**	
	Commiterpenes **A**	**28**	
	Commiterpenes **B**	**29**	
	Commiterpenes **C**	**30**	
	D-Galactose	**31**	
	L-Arabinose	**32**	
	4-Methyl D-glucuronic acid	**33**	
	6-O-(4-O-Methyl-β-D-glucuronosyl)-D-galactose	**34**	
	4-O-(4-O-Methyl-α-D-glucuronosyl)-D-galactose	**35**	
	2-Methoxy-8,12-epoxygermacra-1(10),7,11-trine-6-one	**36**	
	Docosanoic acid	**37**	
	Myrrhone	**38**	
	Mansumbinone	**39**	
Nigella sativa	cis-4-Methoxythujane	**40**	[44,45,47,48,50,51]
	trans-4-Methoxythujane	**41**	
	Thymoquinone	**42**	
	Dithymoquinone	**43**	
	Thymol	**44**	
	Thymohydroquinone	**45**	
	Nigellicine	**46**	
	Nigellidine	**47**	
	Nigellimine	**48**	
	Nigellimine N-oxide	**49**	
	α-Heredin	**50**	
	Quercetin	**26**	
	Kaempferol-3-glucosyl	**51**	
	Quercitin–3-(6-ferulolyl glucosyl)	**52**	
	Triglycoside quercetin 3-glucoside	**53**	
	Kaempferol 3-glucoside	**54**	
	Rutin	**55**	

Ocimum lamiifolium	Bornyl acetate	**56**	[61,68-70]
	p-Cymene	**57**	
	α-Pinene	**58**	
	β-Pinene	**5**	
	Camphene	**59**	
	Sabinene	**60**	
	α-Phellandrene	**61**	
	Apigenin	**62**	
	Genkwanin	**63**	
	Acacetin	**64**	
	Apigenin 7,4'-dimethyl ether	**65**	
	Luteolin	**66**	
	Ladanein	**67**	
	5,6-Dihydroxy-7,3',4'-trimethoxyflavone	**68**	
	5,7-Dihydroxy-6,4'-dimethoxyflavone	**69**	
	Cirsimaritin	**70**	
	Salvigenin	**71**	
	Cirsiliol	**72**	
	Cirsilineol	**73**	
	Eupatorin	**74**	
	5-Hydroxy-6,7,3',4'-tetramethoxyflavone	**75**	
	Quercetin3-O-xylosyl(1'''→2'') galactoside	**76**	
	Flavone 5-O-glycosides	**77**	
Olea europaea	α-Pinene	**58**	[73,77,79,80]
	2,6-Dimethyloctane	**78**	
	2-Methoxy-3-isopropylpyrazine	**79**	
	Hydroxytyrosol	**80**	
	Oleuropein	**81**	
	Verbascoside	**82**	
	Rutin	**55**	
	Luteolin-7-Glucoside	**83**	
	Nüzhenide	**84**	
	Oleuropein Glucoside	**85**	
	Maslinic acid	**86**	
	Oleanolic acid	**87**	
	Ursolic Acid	**88**	
	Erythrodiol	**89**	
	Uvaol	**90**	

Table 1: Lists of compounds that are exist in the five medicinal plants.

inhibited cell proliferation of human ovarian cancer and endometrial carcinoma cells. The inhibitory effects of these solvent extracts (85% EtOH and petroleum ether) on human ovarian cancer cells were strong and observed IC_{50} as 15.8 and 26.91 μg/mL, respectively. Similarly, the IC_{50} values of these extracts on endometrial carcinoma cells were 20.73 and 26.63 μg/mL, respectively [18]. Another biological activity that was investigated on MeOH extract of this plant has shown antifungal activity against *Aspergillus niger* and antibacterial activity at higher concentration. However, the petroleum ether extract has strongly shown antibacterial activity against *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa* and antifungal activity to *Aspergillus niger* and *Candida albicans* [24,26]. Moreover, the resin powder extracts of *C. myrrha* were tested against different gram negative bacteria by disc diffusion method and it has been found that EtOAc and hexane extracts to possess bacterial growth inhibition [26]. In addition, the anti-inflammatory test of EtOH extract significantly inhibited the development of paw swelling induced by formalin and significantly reduced acetic acid-induced writhing. Furthermore, in comparison of the pharmacological activities, the petroleum ether fraction was stronger than EtOH extract. This is because the petroleum ether fraction showed significant analgesic activity in the model at the dose of 100 mg/kg ($p<0.01$) and the ethyl acetate fraction exhibited less analgesic activity ($p<0.05$) [19].

Volatile compounds

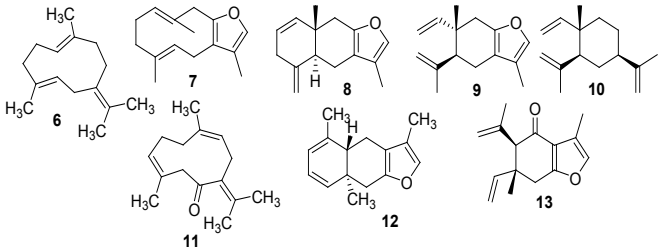
The essential oils of *Commiphora spp.* are rich in furanosesquiterpenoids, which have been found to possess anesthetic, antibacterial, antifungal, antihyperglycemic properties, asthma, bronchitis, for mature and wrinkled skin, gargle for mouth ulcers and thrush and cleanser for the womb, prevention of tooth decay, for treating cancer sores, sore throat, for treatment of mild inflammations of oral and pharyngeal mucosa [21,24].

The minimum inhibitory concentration (MIC) of the essential oil of *C. myrrha* against the toxigenic strain of *Aspergillus flavus* was shown at

3.0 µl/mL concentration [27]. In Somalia the quenching activity against singlet oxygen, an actor of lipid peroxidation and DNA degradation, of the essential oil and resinoid of *C. myrrha* has been studied and compared to DL-α-tocopherol using 1,3-diphenylisobenzofuran (DPBF) as a probe. This research revealed the essential oil and menthofuran has a higher quenching activity than DL-α-tocopherol [25].

Volatile compounds from the oleo-gum resin of *C. myrrha*, which were collected from Ethiopia, were isolated by supercritical extraction method with CO_2 and hydrodistillation. The main components are identified and quantified by GC/MS and has shown the following compounds germacrene B 4.3% (**6**), furanodiene 19.7% (**7**), lindestrene, 12.9% (**8**), curzerene, 8.5% (**9**), β-elemene 8.7% (**10**), germacrone, 5.8%(**11**), and furanoeudesma-1,3-diene 34.9% (**12**) [21,28,29] (Table 1). The irritant potentials of essential oil and seven sesquiterpenoid compounds from this oleo-gum-resin were investigated by open mouse ear assay. The essential oil curzerenone (**13**), furanodiene-6-one (**14**) and furanoeudesma-1,3-diene (**12**) showed potent and persistent irritant effects while others possess least irritant potentials [30].

Another research on the antioxidant and antimicrobial potential of MeOH and EtOAc crude extracts and the essential oil of *C. myrrha* resin were also investigated. As the research revealed the MeOH extract exhibited the highest antioxidant and antimicrobial activity as compared to EtOAc extract and essential oil [31].

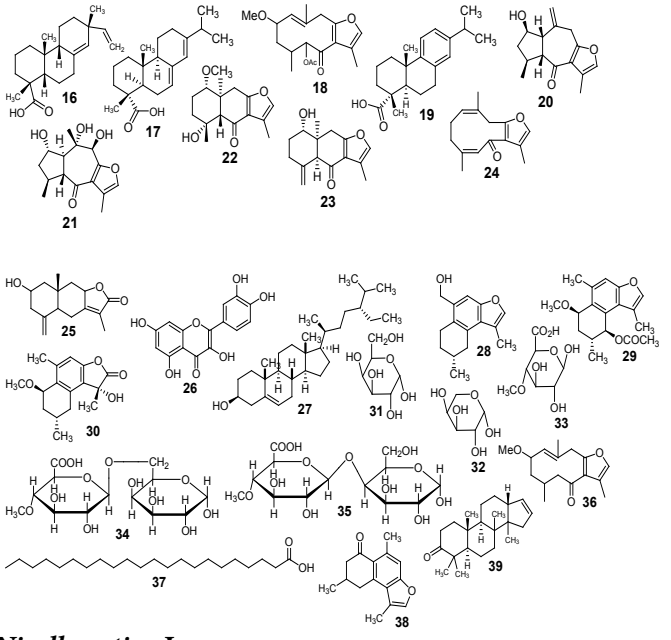


Phytochemicals

Investigation on the resin a new cycloartane type triterpene named cycloartane-1α,2α,3β,25-tetraol (neomyrrhaol) (**15**), along with four known terpenes, sandaracopimaric acid (**16**), abietic acid (**17**), 2-methoxy-5-acetoxyfruranogermacr-1(10)-en-6-one (**18**), and dehydroabietic acid (**19**) have been isolated. Compounds (**18**) and (**19**) exhibited significant aromatase inhibiting activity with IC_{50} values at 0.2 microM and 0.3 microM, respectively. However, compounds (**15-19**) did not inhibit contraction of the isolated uterine [32]. Compounds **16**, **17, 18** and **19** from petroleum ether extract in the concentration rage of 6.25-100 µM were determined about their antiprolifirative activity on various cancer cell lines. The results elucidated that compounds **17** and **19** significantly inhibited cancer cells proliferation of human ovarian cancer cells and cervical carcinoma cells. Compound **19** also significantly inhibited endometrial carcinoma cells. In another comparison while compound **19** inhibited human ovarian cancer cell growth with IC_{50} of 26.93 µM, compound **17** significantly inhibited with IC_{50} of 46.89 µM. This indicates compound **19** has strong effect than **17**. However, it is clear that the two compounds showed remarkably antiproliferative activity on human ovarian cancer cells [18].

In the investigation of the gum exudates and resin of *C. myrrha* has led in the isolation of six sesquiterpenoids Myrrhterpenoid K (**20**), Myrrhterpenoid L (**21**), Myrrhterpenoid M (**22**), Myrrhterpenoid N (**23**), furanogermacra-1(10)*Z*,4*Z*-dien-6-one (**24**) and curzerenone (**13**). Compound (**20**) exhibited weak cytotoxic activity against breast tumor cell line in a clonogenic assay while the other five compounds were inactive [28,33] (Table 1). A sesquiterpen which is commiferin (**25**) was also isolated by another researcher [29].

A further phytochemical investigation afforded quercetin (**26**) and β-sitosterol (**27**) from the aqueous and petroleum ether extracts, respectively [34]. Three cadinane sesquiterpenes, commiterpenes A (**28**), commiterpenes B (**29**), commiterpenes C (**30**) were also isolated from the resinous exudates of *C. myrrha*. All these sesquiterpenes showed neuroprotective effects [28,20]. Extractions with 90% EtOH the resins are largely removed and crude polysaccharides are obtained. After hydrolysis 15 amino acids were detected and in the fractions D-galactose (**31**), L-arabinose (**32**), and 4-methyl D-glucuronic acid (**33**) (in proportions 4:1:3) were identified [29]. Hydrolysis of purified polysaccharides of gum myrrh gave high yields of a mixture of neutral sugars and acidic oligosaccharides. The latter after isolation and purification gave 1:6 mixture of two aldobiuronic acids and identified as 6-O-(4-O-methyl-β-D-glucuronosyl)-D-galactose (**34**) and 4-O-(4-O-methyl-α-D-glucuronosyl)-D-galactose (**35**) [29]. The UPLC–MS/MS chromatogram analysis of petroleum ether extract identified the following compounds 2-methoxy-8,12-epoxygermacra-1,7,11-trine-6-one (**36**), docosanoic acid (**37**), myrrhone (**38**) and mansumbinone (**39**) [18].



Nigella sativa L.

The genus *Nigella* belongs to Ranunculaceae family which contains about 20 species. These species are mostly occurring in Europe, the Mediterranean, central Asia and Africa. Even though there are many species in the genus *Nigella*, in Ethiopia only one species (*Nigella sativa* L.) is widely cultivated and the others are an ornamental. *N. sativa* is also cultivated and have been used to treat various diseases in other parts of Africa such as in Kenya, Somalia, Djibouti, Sudan, northern Africa, and outside Africa in Afghanistan, central and southern Europe, India, Iran, Syria, the Soviet Union and other countries [35]. The seed of this plant has various names as black seed, black cumin, black caraway, funnel flower, roman coriander and kalunji. It is found in most local markets and assumed to be grown through-out the agricultural highlands. The local name of this plant in Ethiopia is called ''*Tikur azmmud*'' [17].

N. sativa is annual herbs and usually profusely branched herb up to 0.7 m tall with finely divided leaves. The leaf segments narrowly linear to threadlike. The stem is ribbed, sometimes hollow when old. The flowers are pale blue or pale purple, with 5-10 petals. The fruit is a capsule composed of several united follicles, each containing numerous seeds [35,36]. Its seeds are small, black and possess aromatic odor and taste [37]. It is an indigenous plant to Ethiopia and most other countries. It is a cultivated plant mostly grown in back gardens of homesteads, on heavier soils as a field crop, tolerating a wide range of soil types, occasionally found and growing wild between 1500-2500 m height [35].

Medicinal uses

N. sativa is an important spice in the culinary to add as flavor. The main uses of this seeds in Ethiopia and other countries are as bread flavor, to prepare hot pepper sauce and dishes. It is mixed with melted butter, wrapped in a piece of cloth and sniffed to relieve some types of headaches. Medicinally it has been used in the folk practice as a natural remedy for a number of diseases such as asthma, hypertension, diabetes, inflammation, cough, eczema, fever and gastrointestinal disturbances. Its seed oil has also antipyretic, analgesic and antineoplastic activity. In Ethiopia it is used to treat headache, stomachache and to induce abortion [35,37]. Bhatti et al. [38] reported that *N. sativa* is a common drug used in Tibbe-Nabvi (Prophet's Medicine) throughout the world because of its therapeutic efficacy and potential to cure disease. Even though *N. sativa* L. used as medicine in different parts of the country, the DNA fingerprints of four Taxa of black seeds from Qassim (Saudi Arabia), Ethiopia, Egypt, and Syria are not identical. The study revealed there are several genetic differences in black seeds Taxa among in these countries [39].

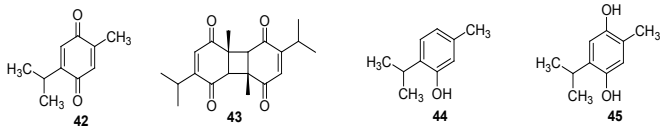
There are ample of scientific evidences for its hypoglycemic, antioxidant, liver protecting, analgesic, antithrombotic, spasmolytic, anti-tumor and antibacterial effects. For example, the research that was done based on identification of the effective of extraction method; the seed was extracted by supercritical carbon dioxide ($SCCO_2$) and Soxhlet extraction using various organic solvents. The antibacterial activities of these extracts are investigated by the agar dilution method against gram-positive bacteria (*Bacillus cereus* and *Staphylococcus aureus*) and gram-negative bacteria (*Escherichia coli* and *Yersinia enterocolitica*). As the result revealed the $SCCO_2$ (at 120 bar/40°C) extract showed effective growth inhibition than solvent extracts against in all the tested bacteria [37]. The result of this investigation has been supported by other research using organic solvent. The MeOH extract exhibited a significant antibacterial activity at the concentration of 50 mg/mL ($p \leq 0.01$) against gram-positive bacteria (*Streptococcus pyogenes*) with a greater inhibition zone of 19 mm than gram negative bacteria (*Pseudomonas aeruginosa*, *Klebseilla pneumonia* and *Proteus vulgaris*) that has 15 mm zone of inhibition. However, *Klebseilla pneumonia* and *Proteus vulgaris* showed resistance against aqueous extract at 20 mg/mL [40]. The ethanol extract of the seed part can also generate antioxidants, possess antitumor activity, and ameliorate and prolong the life span of mice bearing Ehrlich ascites tumor [41].

The study that was conducted to evaluate the *in vitro* and *in vivo* anti-cancer effect of *N. sativa* seed extracts, the essential oil (IC_{50}=0.6%, v/v) and EtOAc (IC_{50}=0.75%) extracts were more cytotoxic against the murine mastocytoma cell line than the butanol extract (IC_{50}=2%). Similar results were also obtained with the kidney carcinoma cell lines of monkeys (Vero). Although all extracts had a comparable cytotoxic effect against the sheep heart carcinoma cell line, with IC_{50} values ranging from 0.2 to 0.26% (v/v), tests on the kidney carcinoma cell lines of hamsters revealed a high cytotoxic effect of the EtOAc (IC_{50}=0.2%) as compared to the essential oil (IC_{50}=1.2%). These data show that the cytotoxicity of each extract depends on the tumor cell type [42]. The powder of seed of *N. sativa* was also investigated *in vivo* by oral administration to the hypercholesterolemic patients (n=10) at dose of 1 g before breakfast for two months to observe its activity. The result revealed it reduces total cholesterol (p=0.002), triglyceride (p=0.0001), HDL-cholesterol (p=0.0003) and LDL-cholesterol level (p=0.002) [38].

In the determination of the toxic effect of *N. sativa* seed powder on the liver function of rats which was evaluated by measuring liver enzymes and through histopathological examination of liver tissue with supplementation up to the dose of 1 g/kg for a period of 28 days, resulted with no changes in liver enzymes level and did not cause any toxicity effect on the liver function [43].

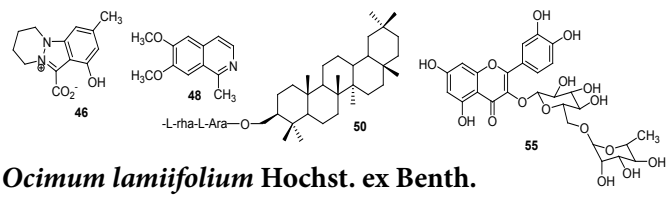
Volatile compounds

The seed part of *N. sativa* contains up to 40% oil and is used in medicine and perfume industry [17]. Using GC/MS instrument forty-eight compounds have been identified in the essential oil from seed part which is cultivated in Poland. The two monoterpenoids *cis*-4-methoxythujane (**40**) and *trans*-4-methoxythujane (**41**) occur as 0.3% and 4%, respectively [44]. Even though in another GC/MS analysis of the volatile oils gave many compounds, the pharmacologically active constituents are thymoquinone (**42**), dithymoquinone (**43**), thymol (**44**) and thymohydroquinone (**45**) [45]. Comparison analysis of solvent extracts was investigated to show their biological activities. As the research revealed the ether and methanol volatile oil extracts have significant antifungal and antibacterial activities as compared to tetracycline, cefuroxime and ciprofloxacin positive controls, the ether extracts showed stronger activity than methanolic one [46].



Phytochemicals

According to phytochemical investigation four alkaloids have been reported as the major constituent of *N. sativa* seeds such as nigellicine (**46**) and nigellidine (**47**) have an indazole nucleus whereas nigellimine (**48**) and nigellimine N-oxide (**49**) are isoquinolines [47-49]. Recently, a triterpene saponin α-Heredin (**50**) was isolated from the seeds of *N. sativa*. α-Heredin is known to have antitumor activity [50]. The ethanolic extract of seed was found to contain three flavonoids namely quercetin (**26**) and kaempferol 3-glucosyl (**51**) and quercitin–3-(6-ferulolyl glucosyl) (**52**) [51]. Other than those triglycoside quercetin 3-glucoside (**53**), kaempferol 3-glucoside (**54**) and rutin (**55**) were also isolated from this seeds [50].



Ocimum lamiifolium Hochst. ex Benth.

The genus *Ocimum* comprises more than 150 species and it is considered as one of the largest genera of the Lamiaceae family [52]. *Ocimum* is a versatile aromatic genus well known for medicinal properties and for economically important essential oils. The genus is very variable and possesses wide range of intra and inter-specific genetic diversity [53].

Ocimum lamiifolium Hochst. ex Benth. is an indigenous plant in Ethiopia and locally is called "*dama-kassie*" (in Amharic) and "*anchabi*" (in Oromifa). It is among the commonly occurring *Ocimum spp.* that distributed in different regions of Ethiopia [54]. It is also cultivated and used as medicine in East Africa from Kenya to Malawi, Dem. Rep. Congo, Cameroon and many other countries [55].

O. lamiifolium is an erect, robust branching sub shrub or shrub growing about 0.7-3 m tall and indumentum of simple hairs. Leaves are ovate and opposite, and flowers are pinkish in racemes. It grows beside roads and streams, in bush land and at forest edged and on grassland between 1200-2900 m [54]. It has petiolate leaves; which is 0.1-0.5 times as long as the blade, up to 40 mm long [54,55].

Ethnobotanical uses

Traditionally the leaves part of *O. lamiifolium* is used against eye diseases and headaches [56]. It is also used to relieve pain and fever in a special local preparation in Ethiopia known as "*Yemich medhanit*" [54,55]. The fresh leaves water extract is drunk to treat diarrhoea, amoeba (diarrhoea with blood) and cough/cold [57]. The plant have synergic activity on these diseases when the water extracts of fresh leaves of *Vernonia amygdalina* and *Clutia abyssinica* mixed with root of *O. lamiifolium* [58]. A research which was done in Sheko ethnic group in Southwest Ethiopia, *O. lamiifolium* is used to treat skin and gastro-intestinal ailments. In this study the plant has been assigned with the highest fidelity level values, a possible indication of their better healing potential [55]. An ethnomedicinal survey that was conducted on the investigation of knowledge and practice of traditional antimalarial plants, the leaf part of *O. lamiifolium* has been traditionally used for antimalarial activity [59]. The thermal expulsion and direct burning of the leaves part revealed significant repellency against the main vectors of malaria [60].

Medicinal uses

As the study about the use of medicinal plants in self-care in rural central Ethiopia revealed, *O. lamiifolium* is among the most frequently used plants [57]. The above ethnobotanical uses of this plant were also corroborated by a scientific laboratory based research. *In vitro* research was done by agar disc diffusion method on the aqueous, ethanol and methanol extracts of *O. lamiifolium*. The result revealed the three extracts have antibacterial activity against *Staphylococcus aureus, Pseudomonas aeruginosa, Escherichia coli* and *Shigella boydii* [61,62]. Evaluation research was done on the antipyretic effects of aqueous and ethanol extracts in mice and the result revealed the extracts have antipyretic property [53]. These extracts were also investigated on anti-inflammatory activities. Both have significant activity but greater anti-inflammatory activity was observed for the aqueous extract. Aqueous extracts of the plant allowed animals to recover barbiturate sleep duration in proportions of 88% [63]. The dried methanolic extract was also tested *in vitro* for its protection activity against acetaminophen-induced hepatotoxicity. The result of this study revealed the plant has activity [64].

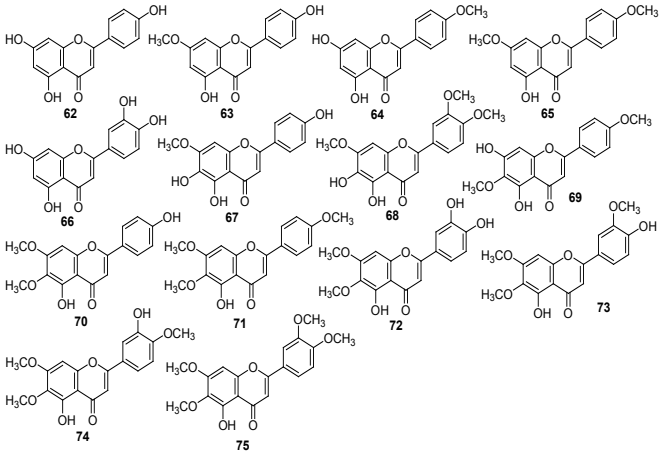
Volatile compounds

Ocimum spp. and their essential oils have been traditionally used to kill or repel insects, and also to flavor foods and oral products, in fragrance, in folk medicine and as condiments [65]. Nerio et al. studied and reported that *Ocimum spp.* have promising essential oils and used as insect repellents [66].

The essential oil of *O. lamiifolium* was analyzed by GC and GC/MS and revealed to have major compounds such as Bornyl acetate (**56**) (30.3%), p-cymene (**57**) (11.4%), α-pinene (**58**) (5.7%), β-pinene (**5**) (4.2%) and camphene (**59**) (5.9%). The oils were also evaluated for antimicrobial activity and have showed moderate activity [67]. The antioxidant activity of essential oils and methanolic extracts of *O. lamiifolium* was evaluated by DPPH and linoleic acid peroxidation assays. As the result revealed sabinene (**60**) (12.2%) and α-phellandrene (**61**) (11.6%) volatile oils scavenge DPPH and inhibit linoleic acid peroxidation. This inhibition property suggested that the plant has hepatoprotective activity [68].

Phytochemicals

The phytochemical compounds are investigated by HPLC, APCI-MS, UV and NMR method and the following flavonoid compounds are isolated and characterized: apigenin (**62**), genkwanin (**63**), acacetin (**64**), apigenin 7,4'-dimethyl ether (**65**), luteolin (**66**), ladanein (**67**), 5,6-dihydroxy-7,3',4'-trimethoxyflavone (**68**), 5,7-dihydroxy-6,4'-dimethoxyflavone (**69**), cirsimaritin (**70**), salvigenin (**71**), cirsiliol (**72**), cirsilineol (**73**), eupatorin (**74**), 5-hydroxy-6,7,3',4'-tetramethoxyflavone (**75**) [69]. Using NMR spectroscopy quercetin 3-O-xylosyl(1'''→2'') galactoside (**76**) was characterised from the leaf extract. *O. lamiifolium* produced larger amounts of flavone 5-O-glycosides (**77**) [70].



Olea europaea L.

The genus *Olea* belongs to Oleaceae family which comprises about 30 species. Olive (*Olea europaea* L.) is one of the species and native to tropical and warm temperate regions of the world. The tree is usually distributed in many parts of the world and used as table oil in the coastal regions of the Eastern Mediterranean Basin, the neighboring coastal areas of south Eastern Europe, western Asia, northern Africa, Tunisia, California, Chile, Croatia, Argentina, South Africa, Saudi Arabia, Australia as well as northern Iran at the south end of the Caspian and other countries. *O. europaea* is an indigenous plant to Ethiopia and locally known as "*Woira*" which yield up to 40% olive oil. The fruit of this wild African olive tree is small and the yield of its oil is low. It has been declined in abundance due to over-utilization and exploitation that is not accompanied by a sound conservation strategy [71].

O. europaea is a small tree growing up to 5-15 m. It grows between the altitudes of 800-2500 m. Bark is grey to brownish-blackish, smooth to rough with old. Leaves are narrowly oblong-elliptic, 2-10 cm × 7-17 mm, grey-green to shiny dark green above, greyish or yellowish with a dense covering of silvery, golden or brown scales on the under surface; apex and base narrowly tapering, apex sharp tipped; margin entire,

rolled under and curved back from the midrib, petiole slender, up to 10 mm long, so the leaves tend to droop. Flowers greenish-white or cream, 6-10 mm long, sweetly scented, in loose axillary or occasionally terminal heads, 5-6 cm long [72].

Medicinal uses

O. europaea is a highly valued plant and its leaves, twigs and woods are used to fumigate pots for milk and for the local beverages in Ethiopia known as "*Tella*" and "*Tej*". Twigs are used as toothbrushes (tooth sticks) and hard wood for carving. The roots are used to treat hemorrhoids. More uses are for preventing heart disease, for treating gout, malaria, stomach problems, hemorrhoids, wrinkles, and to treat malaria [73].

Alcoholic olive leaves extract revealed antibacterial effect [24]. Methanol and chloroform extracts of the leaves part have dose-dependent anti-inflammatory and antinociceptive activities [74]. Another research revealed using disc diffusion and microdilution methods the antimicrobial activity of olive leaf extracts was screened against gram-positive and gram-negative bacteria. According to this study the extract revealed antibacterial activity against *Listeria monocytogenes*, *Bacillus cereus*, *Staphylococcus aureus*, *Enterococcus faecalis*, *Proteus vulgaris*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Enterobacter sakazakii*, and *Escherichia coli*. However, the extract has low antibacterial activity against *Salmonella typhimurium* [75]. The olive leaf is useful as an antioxidant [76].

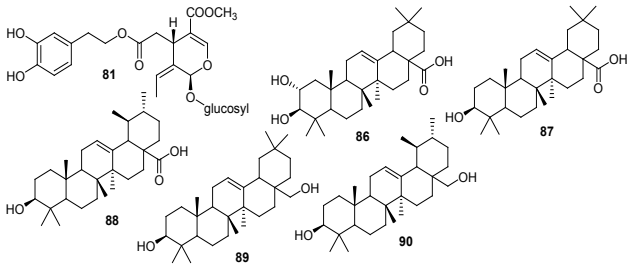
Volatile components

GC and GC-MS analysis of the essential oil resulted in the identification of 32 compounds representing 99.44% of the oil; α-pinene (**58**) (52.70%), 2,6-dimethyloctane (**78**) (16.57%) and 2-methoxy-3-isopropylpyrazine (**79**) (6.01%) were the main components. This essential oil showed radical scavenging, anti-inflammatory and analgesic activities [77].

Phytochemicals

Hydroxytyrosol (**80**) is mainly found in olive leaves after hydrolysis of oleuropein (**81**) and has antioxidant, antibacterial and anti-inflammatory properties. An investigation was done to see the effect of hydroxytyrosol for alleviating the pain in patients with gonarthrosis. The result revealed administered hydroxytyrosol has significant improvement [78]. Oleuropein is the major active compound in olive leaf and well known for its benefits for human health. It is considered as premier cooking oil, used for body care. The research study revealed oleuropein (**81**) is found in the leaves and has significant antispasmodic, antioxidant, diuretic and lipid lowering properties [73]. Extracts of leaves, fruits and seeds of olive tree were analysed by reverse phase HPLC with diode array detection and mass spectrometry. This methodology allowed the identification of verbascoside (**82**), rutin (**55**), luteolin-7-glucoside (**83**), oleuropein (**81**), nüzhenide (**84**), oleuropein glucoside (**85**) and hydroxytyrosol (**80**) [79].

Pentacyclic triterpenes from *O. europaea* are gaining interest due to their beneficial health effects, as anti-inflammatory, anti-diabetic and anti-tumoral. By LC–MS, APCI and LC-MS instruments the following triterpenes: maslinic (**86**), oleanolic (**87**) and ursolic acids (**88**) along with erythrodiol (**89**) and uvaol (**90**) were identified. These compounds are the main triterpenes compounds that present in the fruits and leaves of *O. europaea* [80-82].



Conclusion

Nowadays, many medicines that have been widely in used come from plant resources. Traditionally, the vast majority of people rely on traditional plant based medicines for primary health care [3]. There are many indigenous plants in the world which have high medicinal activities. To mention some of them *A. corrorima*, *C. myrrha*, *N. sativa*, *O. lamiifolium*, and *O. europaea* are among very well known plants in serving the people as spice, flavor, fumigants, and traditional medicines. Some of their biological activities have been corroborated by scientific researches and have shown antibacterial, antioxidant, antifungal, antimicrobial and anti-inflammatory activities. Based on the literature review, majority of the biological researches on these plants relied on *in vitro* analysis which is pivotal as precursor for further investigation. However, it is pertinent further to investigate on *in vivo* and animal model about their pharmacological and pharmacokinetic activities. This is because by sticking only *in vitro* analysis we cannot be sure about their potent effect on metabolic pathway. Some *in vitro* active plant extract may not be active *in vivo* and animal model because the nature of some active compounds is being metabolized.

The phytochemical investigation also revealed these plants are holder of copious kinds of active compound classes and showed wide spectrum of biological activities. To point out, *A. corrorima* have terpene compounds; *C. myrrha* holder of terpenoid, steroid, flavonoid, glycoside and fatty acid. In *N. sativa* terpenoid, steroid, alkaloid compounds are present. *O. lamiifolium* also afforded in extraction terpenoid and flavonoid compounds while *O. europaea* gave terpenoid and steroidal compounds (Table 1). Hence, from these all kinds of compound classes, terpene compound class is a common type among these five medicinal plants. Moreover, even though each plant gives different kinds of active compounds, there are some chemical compounds that are co-exist in one another in a different proportion such as compound **5** is found in *O. lamiifolium* in 4.2% whereas in *A. corrorima* it is found in a larger amount 17.6%. Compound **58** is found in *O. europaea* in larger amount 52.70% than *O. lamiifolium* 5.70%. Compound **26** is also found in both *N. sativa* and *C. myrrha*. Compound **55** is also found in *O. europaea* and *N. sativa* (Table 1). Therefore, from the point of view of ethno-medicinal usage, still it needs further investigations on the isolation of compounds and cross checking of their pharmacology and pharmacokinetics activity on *in vivo* and model to ascertain those traditional practices are really true and also to see whether these compounds have synergic or additive effects when the local people use these plants as a blend form with other medicinal plants.

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Citation: Getasetegn M, Tefera Y (2016) Biological Activities and Valuable Compounds from Five Medicinal Plants. Nat Prod Chem Res 4: 220. doi:10.4172/2329-6836.1000220