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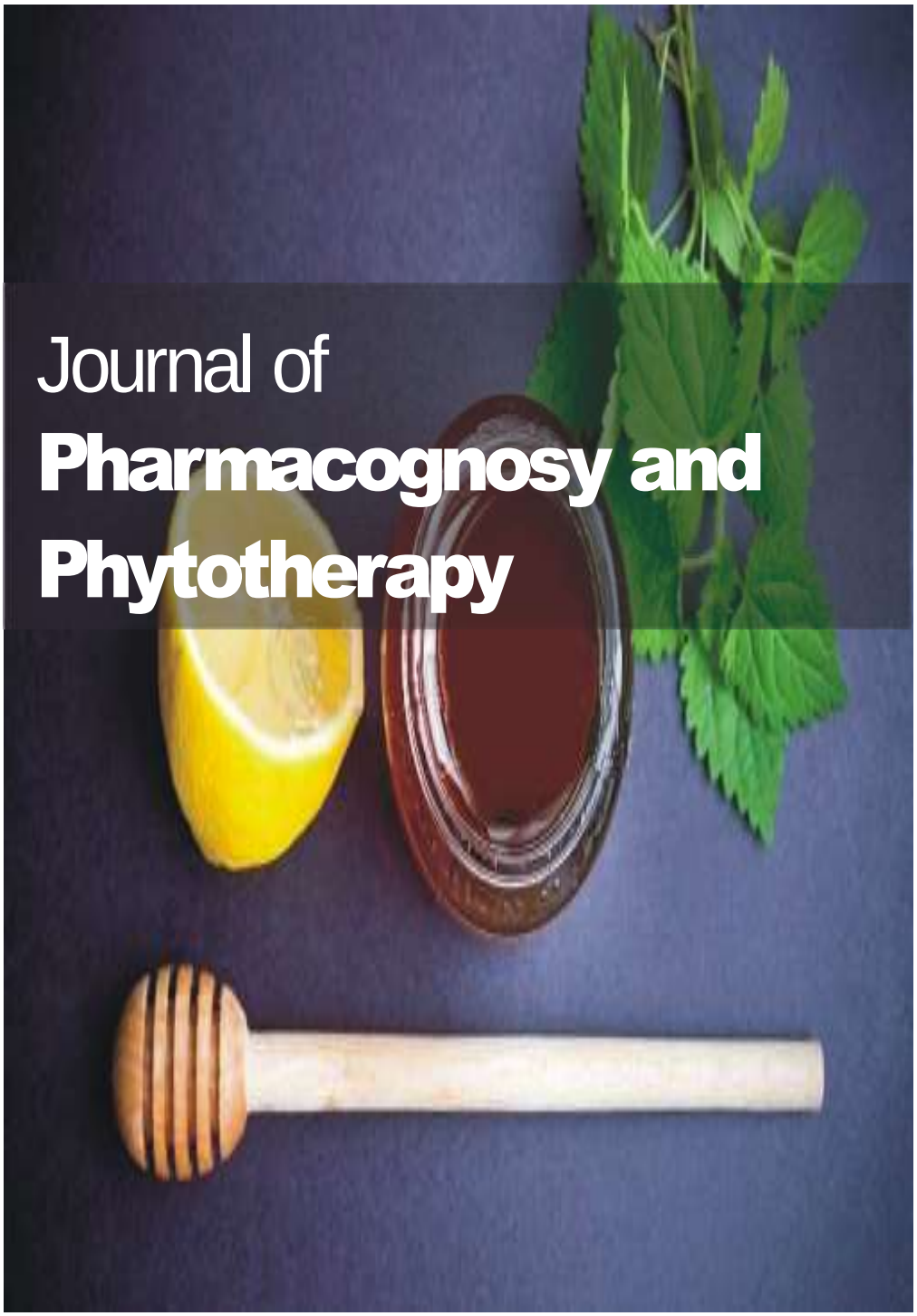

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Pharmacognosy and Phytotherapy

Full Length Research Paper

Phytochemical investigation and assessment of antimicrobial, anti-inflammatory and antioxidant activities of Sudanese *Citrus paradisi* peel extract

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The peel of grape fruit is used in traditional medicine for the treatment of several diseases. The objective of this study was to evaluate the antimicrobial, anti-inflammatory and antioxidant activities of peel extract from *Citrus paradisi.* Qualitative phytochemical screening of peel indicates the presence of alkaloids, flavonoids, sterols, triterpenes, tannins, saponins, coumarins, glycosides, reducing sugars, anthraquinones, lignin and carbohydrates. Extract was assessed for their effectiveness against four bacterial strains including both Gram-positive (*Bacillus subtilis* and *Staphylococcus aureus*) and Gram-negative (*Escherichia coli* and *Salmonella typhi*) bacteria as well as fungal species (*Candida albicans* and *Aspergillus niger*) using disc diffusion method. Antibacterial effects of peel extract showed different degrees of inhibition profiles against tested bacteria with inhibition zone that ranged from 13 to 17 mm. Peel extract showed high antifungal activity against *A. niger* (24 mm) and *C. albicans* (22 mm). The *C. paradisi* peel showed high anti-inflammatory activity with inhibition percentage 77.57%. The antioxidant potential of extract was determined on the basis of their scavenging activity of the 1,1-diphenyl-2-picryl hydrazyl (DPPH) free radical stability. The peel extract showed DPPH scavenging activity (55%) and vitamin C content was 23.08 mg/kg by HPLC. The quantitative analysis of chemical composition of the extract was determined by Gas Chromatography–Mass Spectrometry (GC-MS). The results showed high amounts of Naringenin (28.09%). The peel extract of *C. paradisi* is a natural source of chemical constituents which have medicinal uses in treating many disease.

Key words: *Citrus paradisi* peel, antimicrobial activity, phytochemical screening, vitamin C, Naringenin.

INTRODUCTION

Citrus is one of the most consumed fruits in the world and contain a high amount of useful by-products which

include essential oils. It is mostly consumed fresh or used as raw materials for juice and wine. The second largest world produced citrus species is grape fruit, with an average of more than 60 million annual production. Grapefruit (*Citrus paradisi*) belongs to the family Rutaceae. The yield of grapefruit and oranges juice is about half of the fruit weight thereby generating a very high amount of waste annually (Okunowo et al., 2013). It has been used as a folk medicine in many countries as antibacterial, anti-fungal, anti- inflammatory, antimicrobial, antioxidant, antiviral, astringent, and preservative. It has also been used for cancer prevention, cellular regeneration, lowering of cholesterol, cleansing, detoxification, heart health maintenance, Lupus nephritis, rheumatoid arthritis and weight loss.

In Sudan, *C .paradisi* fruit peel is used for treatment of malaria, gastro protective and antiulcer and this action is attributed to the antioxidant activity of citrus flavonoids found in grapefruit such as naringenin. The major flavonoid exhibited the potent antibacterial and anti *helicobacter pylori* activity *in vitro* and was also recently implicated in cytoprotection against injury induced by algal toxins in isolated hepatocytes. Moreover naringenin showed gastro protective activity due to increased expression of prostaglandins biosynthesis. Furthermore, it was shown to exhibit anticancer activity against human breast cancers. Therapeutic efficacy of citrus fruits such as red grapes and grapefruits is emphasized by the fact that they contain different classes of polyphenolic flavonoids, which were shown to inhibit platelet aggregation thus decreasing the risk of coronary thrombosis and myocardial infarction (Gupta et al., 2011).

An important component of *C. paradisi* is vitamin C. It is an essential micronutrient for humans, with pleiotropic functions related to its ability to donate electrons and a potent antioxidant and a cofactor for a family of biosynthetic and gene regulatory enzymes. Vitamin C contributes to immune defense by supporting various cellular functions of both the innate and adaptive immune system (Traber and Stevens, 2011). It supports epithelial barrier function against pathogens and promotes the oxidant scavenging activity of the skin, thereby potentially protecting against environmental oxidative stress. Vitamin C deficiency results in impaired immunity and higher susceptibility to infections. Furthermore, supplementation with vitamin C appears to be able to both prevent and treat respiratory and systemic infections. Prophylactic prevention of infection requires dietary vitamin C intakes that provide at least adequate, if not saturating plasma levels (that is 100 to 200 mg/day), which optimize cell and tissue levels (Carr and Maggini, 2017).

In the present paper, results on phytochemical screening of the 96% ethanolic extract of *C. paradisi* fruits peel and assessment of it's antimicrobial, anti-inflammatory and antioxidant activities in addition to determination of vitamin C and naringenin content by HPLC and GC-MS analysis was reported.

MATERIAL AND METHODS

Preparation of peel extract

The peel of fresh fruit of *C. paradisi* was air dried and ground to powder using a pestle and mortar. A hundred grams of powder was extracted with 96% ethanol at room temperature for 72 h. The extract was first filtered through Whatman number 4 filter paper. After filtration, the extract was vacuum concentrated.

Phytochemical analysis

Qualitative preliminary phytochemical analysis was performed initially with different chemical reagents to detect the nature of phytoconstituents and their presence in peel. The presence of sterols/terpenes, flavonoids, tannins, alkaloids, lignins, saponins and coumarins were evaluated by standard qualitative methods of Trease and Evans (Trease and Evans, 2002).

Antimicrobial activity

Test microorganisms

Six microorganisms were used in this study, consisting of four bacterial strains and two fungal strains. Two were Gram positive (*Bacillus subtilis* and *Staphylococcus aureus*), while the other two were Gram negative (*Escherichia coli*, and *Salmonella typhi*). The two fungal strains used were *Candida albicans*, *Aspergillus niger*. Standard strains of microorganism used in this study were obtained from Medicinal and Aromatic Institute of Research, National Research Center, Khartoum.

Culture media

Mueller Hinton agar

Thirty eight grams of the powder of Mueller Hinton agar were weighed, dissolved in 1 liter of distilled water and allowed to soak for 10 min. The medium was placed in water bath to dissolve, swirled to mix and sterilized by autoclaving for 15 min at 121°C, cooled to 47°C mixed well then poured into sterile Petri dishes.

Sabouraud Dextrose agar

Sixty two grams of the powdered Sabouraud dextrose agar, was weighed, dispersed in 1 L water and allowed to soak for 10 min, swirled to mix then sterilized by autoclaving for 15 min at 121°C, cooled to 47°C, mixed well and then poured in to sterile Petri dishes.

Antibacterial assay

The disc-diffusion assay (Kil et al., 2009) with some modifications

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was employed to investigate the inhibition of bacterial growth by peel extract. Extract solution (20 mg/ml) was prepared by diluting with dimethyl sulfoxide (DMSO) 30%. Base plates were prepared by pouring 10 ml Mueller-Hinton (MH) agar into sterile Petri dishes. About 0.1 ml of the standardized bacterial stock suspension 10^8 to 10^9 C.F.U/ ml were streaked on Mueller Hinton agar medium plates using sterile cotton swab. Sterilized filter paper disc (6 mm diameter) were soaked in the prepared extracts, and then were placed on surface of the test bacteria plates. The plates were incubated for 24 h and the diameters of the inhibition zones were measured.

Antifungal assay

The same method described for bacteria was employed to antifungal activity, Sabouraud Dextrose Agar was used. The inoculated medium was incubated at 25˚C for two days for the *Candida albicans* and three days for *Aspergillus niger*.

Anti-inflammatory activity

Inhibition of albumin denaturation

Inhibition of protein denaturation was evaluated by the method of (Sakat et al (2010)) with slight modification: 500 µL of 1% bovine serum albumin was added to 100 µL of plant extract with different concentrations. This mixture was kept at room temperature for 10 min, followed by heating at 51°C for 20 min. The resulting solution was cooled down to room temperature and absorbance was recorded at 660 nm. Standard (Aspirin) was taken as a positive control. The experiment was carried out in triplicates and percent inhibition for protein denaturation was calculated using: % Inhibition = (A0 – A1) /A0 *100
Where, A0 is the absorbance of the control and A1 is the absorbance of the sample.

Antioxidant activity

The DPPH radical scavenging was determined according to the method of Shimada et al (1992), with some modification. In 96-wells plate, the test samples were allowed to react with 2,2, Di (4-tert-octylphenyl)-1-picryl-hydrazyl stable free radical (DPPH) for half an hour .The concentration of DPPH was kept as (300 µl). The test samples were dissolved in DMSO while DPPH was prepared in ethanol. After incubation, decrease in absorbance was measured at 517 nm using Shimadzu UV spectrophotometer double beam. Percentage radical scavenging activity by samples was determined in comparison with DMSO treated control group. Ascorbic acid was used as standard. The ability to scavenge the DPPH radical was calculated using the following equation:

$$\text{DPPH scavenging activity (\%)} = (A0 - A1)/A0 \times 100$$

Where, A0 is the absorbance of the control and A1 is the absorbance of the sample.

Quantitative estimation by HPLC to determine ascorbic acid (vitamin C) in peel extract

The HPLC analysis system was Waters 2996 Photodiode array detector and Waters 2695 Separation Module HPLC pump (Waters, Milford, USA). The chromatographic assay was performed on a Intersil ODS-3 column (4.6 mm x 250) reversed phase matrix (5 µm) (Waters) and elution was carried out in a gradient system with acetic acid 0.1% (w/v) methanol (95:5%). UV detector was set at 254 nm and the volume of injection was 20 µl.

GC-MS analysis

The qualitative and quantitative analysis of the sample was carried out by using GC-MS technique model (GC-MS-QP2010-Ultra) from japans 'Simadzu Company, with capillary column (Rtx-5ms-30 m×0.25 mm×0.25 µm).The sample was injected by using split mode, Helium as the carrier gas passed with flow rate 1.61 ml/min. The temperature program was started from 60°C with rate 10°C /min to 300°C as final temperature degree with 2 minhold time: the injection port temperature was 300°C. The ion source temperature was 200°C and the interface temperature was 250°C. The sample was analyzed by using scan mode in the range of m/z 40-500 charges to ratio and the total run time was 26 min .Identification of components for the sample was achieved by comparing their retention times and mass fragmentation patterns with those available in the library from the National Institute of Standards and Technology (NIST).

RESULTS AND DISCUSSION

Qualitative preliminary phytochemical analysis

Qualitative preliminary phytochemical analysis was performed initially with different chemical reagents to detect the nature of phytoconstituents and their presence in peel extract of *C. paradisi*. Phytochemical screening showed that the peel extract was rich in chemical constituents, results are presented in Table 1. Preliminary phytochemical analysis of peel extract of *C. paradisi* revealed presence of flavonoids, sterols, triterpenoids, coumarins, glycosides, reducing sugars and carbohydrates, but alkaloids, tannins, saponins, anthraquinones and lignin were not detected, and might be present in trace undetectable amounts by qualitative methods. These principles have been known for many years to exhibit biological activity, such as effects on the central nervous system, antibacterial, antitumor and anthehelmintic activity (Harborne, 1973). Compared with previous studies, Mathew et al. (2012) reported the presence of flavonoids, alkaloids, steroids, terpenoids, saponins, cardiac glycosides, and reducing sugars.

Generally, phytochemicals are known to confer certain health benefits such as anti-inflammatory, antimicrobial, antihypertensive, and antidiabetic effects (Oikeh et al., 2016; Oikeh et al., 2013).

Antimicrobial activity

The antibacterial activity of the ethanolic extract from peel of *C. paradisi* was determined against the Gram positive *B. subtilis* and *S. typhi* and the Gram negative *E. coli* and *S. aureus* and two fungi; *C. albicans* and *A. niger* using the disc diffusion method. The results are presented in Table 2. Different extracts showed variable activity

Table 1. Preliminary phytochemical screening of peel 96% ethanolic extract of *C. paradisi* fruit.

Test	Specific test	Grape fruit peel
Alkaloids	Wagner's	-ve
	Mayer's	-ve
	Dragendroff's	-ve
Flavonoids	$FeCl_3$	+ve
	Lead acetate	-ve
Sterols	Salkowski	+ve
	Lebermann	+ve
Triterpenes	Salkowski	+ve
	Liebermann	+ve
Tannins	$FecCl_3$	-ve
	Gelatin	-ve
	HNO_3	-ve
	lead acetate	-ve
Saponins	Foam test	-ve
Coumarins	UV lamp	+ve
Glycosides	Keller kiliani	+ve
	Kedd's	-ve
Reducing sugars	Fehling's	+ve
Anthraquinones	Ammonia test 25%	-ve
Lignins	Labat test	-ve
Carbohydrates	Molisch	+ve

against the tested bacteria. Generally, the Gram-positive strains showed higher susceptibility values than the Gram negative strains. The highest antibacterial activity was showed by *C. paradisi* against *B. subtilis* (17 mm) followed with inhibition zone against *S. aureus* (15 mm) , and against *S. typhi* (14 mm), while the lower zones of inhibition was observed in the Gram negative organisms *E. coli* (13 mm). *C. paradisi* extract exhibited high antifungal activity against *C. albicans* and *A. niger* with inhibition zone (22 and 24 mm) respectively (Figure 1).

In vitro anti-inflammatory activity

Protein denaturation is a process in which proteins lose their tertiary structure and secondary structure by application of external stress or compound, such as strong acid or base, a concentrated inorganic salt, an organic solvent or heat. Most biological proteins lose their biological function when denatured. Denaturation of proteins is a well-documented cause of inflammation (Chandra et al., 2012). Results showed in Table 3. The *Citrus paradisi* peel showed high anti-inflammatory activity with inhibition percentage 77.57%. Aspirin a standard anti-inflammation drug showed the maximum inhibition of 88.59%.

Antioxidant activity

The *in vitro* antioxidant activity of the ethanolic extract from peel of *C. paradisi* fruit was assessed by DPPH

Table 2. Antimicrobial activity of *Citrus paradisi* peel extract.

Extract (20 mg/ml)	MDIZ (Mean diameter of growth inhibition zone, mm)					
	Bacteria strain				Fungi strain	
Peel extract	***B. subtilis***	***S. aureus***	***E. coli***	***S. typhi***	***C. albicans***	***A. niger***
	17±1.41	15±0.00	13 ±0.70	14±0.00	22±1.41	24 ±0.0

Interpretation of results: MDIZ* (mm):< 9 mm inactive; 9-12 mm partially active; 13-18 mm active;>18 mm: Very active.

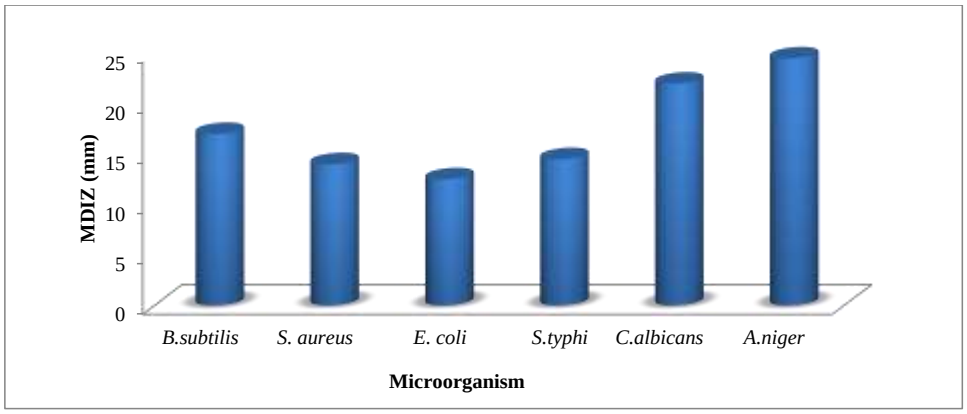

Figure 1. Antimicrobial activity of peel extract against bacteria and fungi microorganisms.

Table 3. Effect of peel ethanolic extract on protein denaturation.

Sample	Inhibition (%)
Grape fruit peel	77.57
Aspirin (Control +)	89.59

Table 4. Antioxidant activity by DPPH assay of *C. paradisi* peel extract.

Sample	DPPH %
Grape fruit peel	55
Ascorbic acid	93.5

assays. Results are shown in Table 4. The extract showed moderate antioxidant activity (55.8 %) compared with ascorbic acid (93.5%). The supplementation of natural antioxidants through a balanced diet containing enough fruits could be much more effective and economical than the use of individual antioxidants, such as vitamin C or vitamin E for protecting of the body against various oxidative stresses (Pisoschi and Pop, 2015).

Barros et al. (2012) stated that antioxidant capacity of all peels was higher than those of pulps, both in terms of the DPPH radical scavenging capacity and the FRAP assay and the antioxidant capacity of citrus does not seem to be a property of a single phytochemical compound, but is correlated both to vitamin C and phenolic constituents.

Determination of vitamin C in *C. paradisi* peel extract by HPLC

C. paradisi peel contains about 23.08 mg/kg of vitamin C (Figure 2 and 3). Previous studies have shown that grape fruit has high vitamin C content and is therefore valuable to the immune system. It helps protect against colds and flu; has a positive effect on obesity and also has diuretic properties. It is used with great success to combat muscle fatigue and stiffness while stimulating the lymphatic system

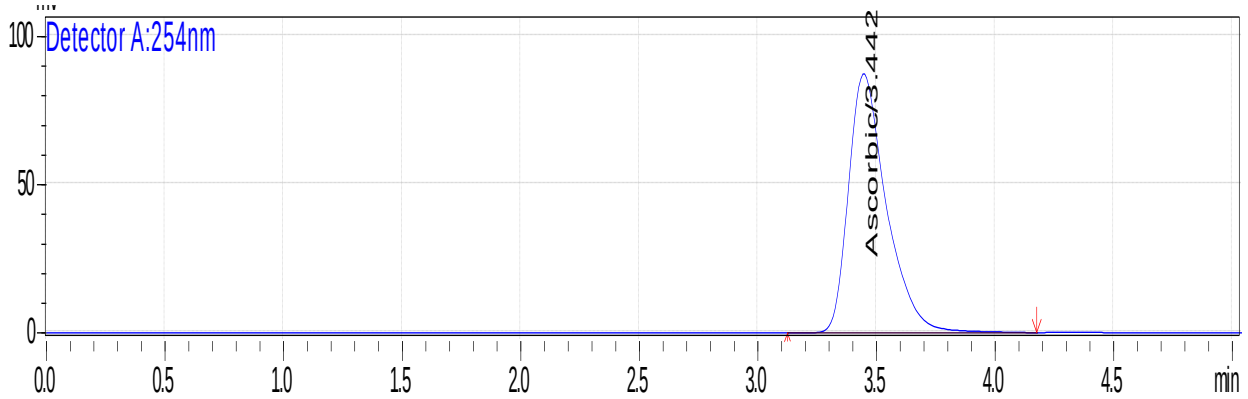

Figure 2. Diagram of vitamin C standard by HPLC.

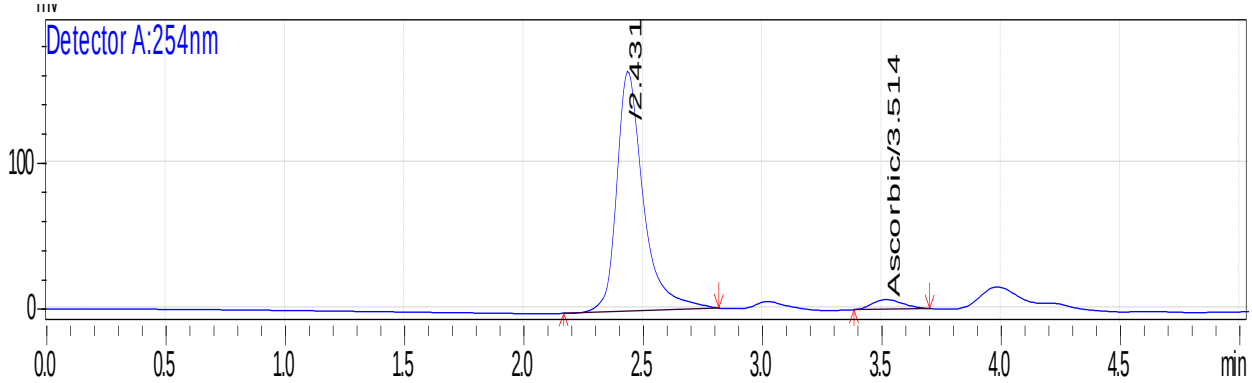

Figure 3. Diagram of vitamin C in *C. paradisi* peel extract by HPLC.

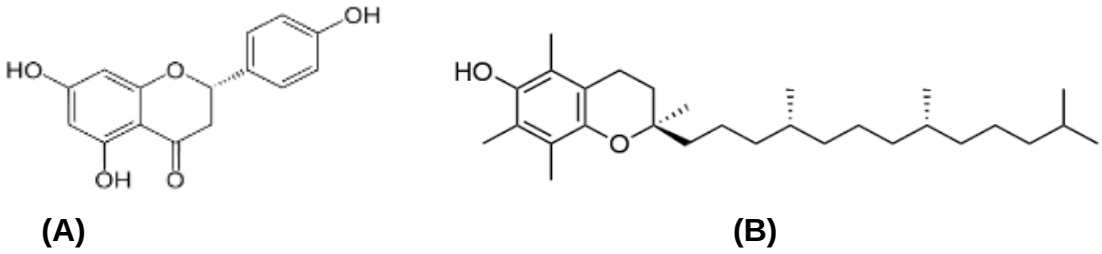

Figure 4. Chemical structure of Naringenin (A) and vitamin E (B).

and therapy clearing the body of toxins (Faleye et al., 2012).

GC-MS analysis

The results of GC-MS analysis of peel ethanolic extract showed different types of chemical constituents (Table 5). The main component in grapefruit peel was found to be Naringenin (28.09%). Citrus flavonoids constitute an important series of flavonoids. Naringenin is a flavanone aglycone of naringin (Figure 4) which has been reported to have numerous bioactive effects on human health such as being an antioxidant, an anti-inflammatory, anti-diabetic and anti-neurodegenerative (Moran et al., 2016).

The results were in agreement with those obtained by Gupta et al. (2011), who reported that, Citrus peel was rich in flavanone glycosides and poly methoxy flavones. Grapefruit peel is candied and is an important source of chemical constituents. Several pharmacological activities

Table 5. Chemical composition of ethanol extract of *C.paradisi* peel using GC-MS.

S/N	R.Time	Name	Formula	Area%
1	4.131	Beta - Myrcene	$C_{10}H_{16}$	0.12
2	4.667	D-Limonene	$C_{10}H_{16}$	0.75
3	5.257	Alpha -Methyl- alpha- [4-mthyl-3-penten]oxiranemethanol	$C_{10}H_{18}O_2$	1.90
4	5.471	Ethyl 2-(5-methyl-5-vinyltetrahydrofuran-2-yl)	$C_{13}H_{22}O_4$	1.64
5	5.590	1,6-Octadien -3-ol,3,7-dimethyl	$C_{10}H_{18}O$	0.46
6	6.373	Ethoxycitronellal	$C_{12}H_{22}O_2$	0.29
7	6.942	L.alpha-Terpineol	$C_{10}H_{18}O$	0.33
8	7.689	(-) Carvone	$C_{10}H_{14}O$	0.06
9	8.012	2-Furanmethanol,5-ethenyltetrahydro-alpha,alpha,5-trimethyl,cis	$C_{10}H_{18}O_2$	0.62
10	8.390	Artemiseole	$C_{10}H_{16}O$	0.17
11	9.446	Geranyl acetate	$C_{12}H_{20}O_2$	0.31
12	9.487	Alpha -Copaene	$C_{15}H_{24}$	0.30
13	10.095	Caryophyllene	$C_{15}H_{24}$	1.30
14	10.538	1,3-Propanediol,2-(hydroxymethyl)-2-nitro methane	$C_4H_9NO_5$	17.94
15	11.068	D-Allose	$C_6H_{12}O_6$	4.82
16	11.343	Naphthalene ,1,2,3,5,6,8a-hexahydro-4,7-dimethyl -1(1-methylethyl)	$C_{15}H_{24}$	0.87
17	11.674	Cyclohexanemethanol ,4-ethenyl-alpha.,4-trimethyl-3-(1-methylethyl)	$C_{15}H_{26}O$	0.59
18	11.724	1,6,10-Dodecatrien-3-ol,3,7,11-trimethyl	$C_{15}H_{26}O$	0.15
19	11.858	3-Oxabicyclo(4.3.0)nonan-2-one,8-isopropylidene	$C_{11}H_{16}O_2$	0.63
20	12.727	1,2,3,5-Cyclohexanetetrol	$C_6H_{12}O_4$	2.23
21	15.647	Benzenemethanol ,alpha-(1-(ethylmethylamino)ethyl	$C_{12}H_{19}NO$	0.44
22	16.003	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	1.14
23	16.031	Isopropyl palmitate	$C_{19}H_{38}O_2$	0.40
24	17.285	7H-Furo(3,2)-(1)benzopyran-7-one,4-methoxy	$C_{12}H_8O_4$	0.56
25	17.722	1(2H)-Naphthalenone,3,4-dihydro-5-methoxy-8-methyl	$C_{12}H_{14}O_2$	2.21
26	17.886	Osthole	$C_{15}H_{16}O_3$	1.92
27	18.375	2,3,5,6-Tetramethylterphthalaldehyde	$C_{12}H_{14}O_2$	0.32
28	18.649	(5,6-Dihydro-2H-(1,4)oxazin-3-yl)-p-tolyl-amine	$C_{11}H_{14}N_2O$	0.54
29	18.806	2H-1-Benzopyran-2-one,7-methoxy-6-(3-methyl-2-oxobutyl)	$C_{15}H_{16}O_4$	3.36
30	18.995	Cholest -5-en-3-ol(3.beta),carbonochloridate	$C_{28}H_{45}CLO_2$	2.82
31	19.227	7-Methoxy-1-methyl-8(1H)-cycloheptapyrazolone	$C_{10}H_{10}N_2O_2$	1.11
32	19.522	Phenacetic amide ,2-methoxy-6-nitrose-alpha,alpha.,dimethyl	$C_{11}H_{14}N_2O_3$	0.51
33	20.443	2,3-Dihydroxydihydrosuberosin	$C_{15}H_{18}O_5$	4.29
34	20.653	Octaethylene glycol monomethyl ether	$C_{19}H_{38}O_{10}$	2.63
35	21.065	Methyl 6-O-(1-methylpropyl).beta-d-galactopyranoside	$C_{11}H_{22}O_6$	1.84
36	21.890	2H-1-Benzopyran-2-one,7(3,7-dimethyl-	$C_{19}H_{22}O_3$	5.34
37	23.158	Isocyclocitral	$C_{10}H_{16}O$	6.41
38	23.997	Naringenin	$C_{15}H_{12}O_5$	28.09
39	25.524	Vitamin E	$C_{29}H_{50}O_2$	0.95

of the peel were reported; anti HIV, anti-inflammatory effect, anti atherogenic, antibacterial, apoptotic activity, anxiolytic, antidepressant and antioxidant.

Conclusion

This study demonstrated support for the claimed uses of the plants in the traditional medicine probably due to the phytochemicals present. The peel of grapefruit is a very important part, as rich source of chemical constituents which is for prevention and cure of diseases. The peel (96% ethanolic extract of *C. paradise*) showed various degree of inhibitory activity against tested microorganism species of bacteria and fungi. Analysis of the peel extract showed high amount of vitamin C and naringenin which might be the cause of the effectiveness against inflammation and antioxidant activity. The results of the present study gave solid grounds that the *C. paradisi* peel extract passes a medicinal potential to develop new phyto-

pharmaceutical drugs and cosmeceuticals.

CONFLICT OF INTERESTS

Authors declare no conflict of interests.

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REFERENCES

Barros HR, Castro Ferreira TA, Genovese MI (2012). Antioxidant capacity and mineral content of pulp and peel from commercial cultivars of citrus from Brazil. Food Chemistry 134:1892-1898.

Carr AC, Maggini S (2017). Vitamin C and Immune Function. Nutrients 9:1211.

Chandra S, Chatterjee P, Dey P, Bhattacharya S (2012). Evaluation of in vitro anti-inflammatory activity of coffee against the denaturation of protein. Asian Pacific Journal of Tropical Biomedicine pp. 178-180.

Faleye FJ, Ogundaini AO , Olugbade AT (2012). Antibacterials and antioxidant activities of Citrus paradisi (Grape fruit seed) Extracts. Journal of Pharmaceutical and Scientific Innovation 1:63-66.

Gupta V, Kohli K, Ghaiye P, Bansal P, Lather A (2011). Pharmacological potentials of Citrus paradisi- an overview. International Journal of Phytothearpy Research 1:8-10.

Harborne JB (1973). Phytochemical Methods, Chapman and Hall, Ltd., London pp. 149-188.

Kil HY, Seong ES, Ghimire BK, Chung IM, Kwon SS, Goh EJ, Heo K, Kim MJ, Lim JD, Lee D, Yu CY (2009). Antioxidant and antimicrobial activities of crude sorghum extract. Food Chemistry 115:1234-1239.

Mathew BB, Jatawa SK, Tiwaari A (2012). Phytochemical analysis of Citrus limonum pulp and peel. International Journal of Pharmacy and Pharmaceutical Sciences 4:269-371.

Moran EP, Wang Z, Chen J, Sapieha P, Smith LE, Ma JX (2016). Neurovascular cross talk in diabeticretinopathy: Pathophysiological roles and therapeutic implications. American Journal of Physiology-Heart and Circulatory Physiology 311:738-749.

Oikeh EI, Omoregie ES, Oviasogie FE, Oriakhi K (2016). Phytochemical, antimicrobial, and antioxidant activities of different citrus juice concentrates. Food science and nutrition 4:103-109.

Oikeh EI, Oriakhi K, Omoregie ES (2013). Proximate analysis and phytochemical screening of Citrus sinensis fruit wastes. Bioscientist 1:164-170.

Okunowo WO, Oyedeji O, Afolabi LO, Matanmi E (2013). Essential Oil of Grape Fruit (Citrus paradisi) Peels and Its Antimicrobial Activities. American Journal of Plant Sciences 4:1-9.

Pisoschi AM, Pop A (2015). The role of antioxidants in the chemistry of oxidative stress: A review. European Journal of Medicinal Chemistry 97:55-74.

Sakat S, Juvekar AR, Gambhire MN (2010). In vitro antioxidant and anti-inflammatory activity of methanol extract of Oxalis corniculata Linn. International Journal of Pharmacy and Pharmaceutical Sciences 2:146-156.

Shimada K, Fujikawa K, Yahara K, Nakamura T (1992). Antioxidative properties of xanthan on the antioxidation of soybean oil in cyclodextrin emulsion. Journal of Agricultural and Food Chemistry 40:945-948.

Traber MG, Stevens JF (2011). Vitamins C and E: beneficial effects fromamechanistic perspective. Free Radical Biology and Medicine 51:1000-1013

Trease GE, Evans WC (2002). Textbook of Pharmacognosy. 15th Ed. Saunders Publishers, London.

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Ethnobotanical survey of plants traditionally used for malaria prevention and treatment in indigenous villages of Tepi Town South West Ethiopia

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Increased resistance to insecticides and established drugs by malaria vectors necessitate the search for alternative cost-effective malaria control tools in the Ethiopia. Traditional remedies are the most important source of therapeutics of the population and more than 85% of the traditional medical preparations in Ethiopia are of plant origin. As the Ethiopian indigenous medicinal plants' knowledge and diversity is vulnerable to be lost continuous documentation and preservation of traditional knowledge and the plant species is a priority. Thus, we report an ethnobotanical survey of plants traditionally used for malaria prevention and treatment in an indigenous villages of Tepi town south western Ethiopia. To document anti-malarial plant traditional knowledge and determine level of utilization for prevention and treatment of malaria by households, 40 household heads were surveyed by snow ball sampling of which eight household heads addressed by systematic purposive sampling were traditional healers. The data were collected through semi-structured interviews and were analyzed using SPSS version 20. A total of twenty five plant species belonging to twenty two families have been reported. The most cited plant species for malaria prevention by healers were *Cyperus* species (52.11%), *Allium sativum* L. (24.15%), *Lepidium sativum* L. (9.34%) and *Echinops kebericho* Mesfin. (7.82%). This study has documented more anti-malarial plant species to be used in the indigenous village. The existing medicinal plant species and the indigenous knowledge on traditional medicinal plants in the study area were under serious threat and were at risk of getting lost. Therefore, urgently warrant sustainable conservation and further research is needed.

Key words: Indigenous knowledge, malaria vectors, medicinal plants.

INTRODUCTION

Malaria is a major public health problem in the tropical part of the world, especially in the sub-Saharan Africa. It is estimated that annually there are 300 million cases of malaria worldwide resulting in one million deaths. Ninety percent of these deaths occur in sub-Saharan Africa, and most of the victims are children under 5 years of age and pregnant women (WHO, 2015). Malaria is caused by five species of parasite that affects humans. All the parasites

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belong to the genus *Plasmodium*: *Plasmodium falciparum, Plasmodium vivax, Plasmodium ovale, Plasmodium malariae,* and *Plasmodium knowlesi*. Of these, *P. vivax* and *P. falciparum* are the most important when it comes to the disease propagation of malaria (WHO, 2013).

Malaria is ranked as the leading communicable disease in Ethiopia; it is a leading cause of outpatient visits (17.0%), inpatient admissions (15.0%), and death (29.0%) in most parts of the country (CSA, 2016). It is estimated that more than 85% of the Ethiopian population does not enjoy the services and benefits of modern medicine (Amare, 1976; Dawit, 1986). Moreover, modern drugs are too expensive for the Ethiopian economy particularly the rural mass.

Increased resistance to insecticides by the major malaria vectors (Balkew et al., 2012, Massebo, 2013) and increased spread of drug resistance by malaria parasites (MOH, 2012). Prohibitive costs of the insecticides and drugs necessitate the search for alternative cost-effective methods for malaria control in Ethiopia (Afrane et al., 2008; Deressa et al., 2006).

In malarial infested regions, affordable treatments against malaria are mainly based on the use of traditional herbal remedies. Indeed, indigenous plants play an important role in the management of the disease, and they seem to be the most convenient solution because of their accessibility and diversity in tropical and sub-tropical regions (Karou et al., 2007; Phillipson and Wright, 1991; Ngutaa et al., 2010; Otten et al., 2009; Alemu et al., 2012).

According to several reports, up to 80% of world's populations still rely on traditional medicine mainly on herbal remedies as primary source of medicinal agents for the treatment of diseases including Malaria (Hostettman and Marston, 2002; Geoffrey and Kirby, 1996). Thus, the development of medicinal plants in primary health care not only will save the foreign exchange but also will aid in conserving our national heritage.

Studies on the anti-malarial plants and their threats in and around Tepi town are limited. Therefore, this study was conducted to document traditional medicinal plant species that are utilized for malaria prevention and treatment and the indigenous knowledge of the people in a native village in south western Ethiopia. Documentation of medicinal plants and the associated indigenous knowledge is very important to conserve the medicinal plant genetic diversity and preserve the knowledge and traditional skills.

MATERIALS AND METHODS

The study area

The study was conducted in an indigenous village of Tepi town Yeki district. The area was purposely selected for this study because it is among the major malaria-prone areas in south west Ethiopia. The villages were selected purposely based on accessibility and knowledge of reported malaria cases. Geographically, the area lies between 7°12' and 7°43' W latitude and 35°32' and 35°75' E longitude found at 611 km far from Addis Ababa.

The town is bounded by Kefa Zone on east, Mejengir Zone of Gambella region on west, Anderacha district on north, and Sheko district of Bench Maji Zone on south. The altitudinal range of the district falls between 1001 and 2007 m above sea level, and it receives high amount of rainfall, with an average of 1171 to 2200 mm annually. Most households primarily depend on subsistence rain fed agriculture and livestock herding. Malaria control heavily relies on long lasting insecticidal nets (LLINs) and by treatment of diagnosed cases with anti-malarial drugs.

An ethno botanical survey

To document an indigenous anti-malarial plant traditional knowledge and determine level of utilization of traditional medicinal plants for prevention and treatment of malaria by households, 40 household heads were surveyed by snow ball (referral) sampling of which 8 household heads were traditional healers included by purposive sampling. The sample household heads from all-age groups were randomly interviewed from the villages. The ethno-botanical techniques employed to collect data on knowledge and usage of medicinal plants was based on semi-structured interviews and field observation. All plant collections were made by the researchers and field assistant who can speak the local language and also familiar with the traditional healers.

For collection of information on plants used for treating and prevention of malaria by the people living in the study villages, a semi-structured questionnaire was prepared that focus to collect socio-economic status of the participants, local names of the anti-malarial plants, plant parts used, and how such knowledge is preserved and transmitted to next generation. With the help of local informants, the plant species were observed in the field and the plant specimens were taxonomically identified with the help of herbarium materials and experts.

Data analysis

Data collected during the survey was checked by the researcher in the field and after data cleaning, coding and editing; entry and analysis was made using the statistical package for social sciences (SPSS) version 20.0 software. Descriptive statistics mainly frequency and cross tab were employed to analyze the proportion of use-reports of medicinal plant species that were frequently cited by the informants during ethno botanical survey.

Ethical considerations

The objectives of the study were clearly explained and verbal consent was obtained from each study participant. Approval to conduct the study was granted by Mizan Tepi University.

RESULTS AND DISCUSSION

Socio-economic characteristics and knowledge of participants on anti-malarial plants

Socio-economic features of respondents are shown in Figure 1. In this ethno-botanical study, majority of the informants who participated in the study were male

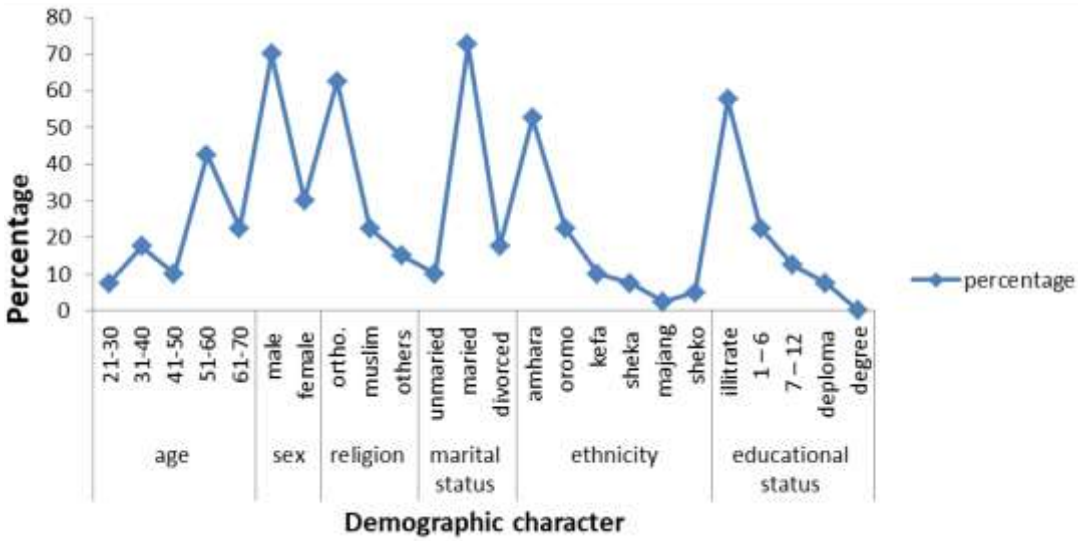

Figure 1. Socio economic characters of informants.

constitute 70% and female were 30% with an age ranging from 21 up to 70. The majority of the informants were illiterate but many of the respondents had adequate knowledge about traditional anti-malarial medicinal plants that are used for malaria prevention and treatment in their localities.

Traditional usage of anti-malarial plants by households

The present ethno-botanical survey results revealed a total of twenty five plant species used by the local community of Tepi town for prevention of malaria (Appendix 1). As it can be seen the most cited and frequently used plant species and its percentage use for malaria prevention in Tepi town by the villages were: *Cyperus* species (52.11%), *Allium sativum* L. (28.15%), *Lepidium sativum* L. (13.34%) and *Echinops kebericho* Mesfin (9.82%).

The 25 plant species that were mentioned by the informants as effective against malaria and their uses that were documented during field survey work are provided. The respondents had good knowledge about malaria and could identify it from other fevers on the basis of locally accepted characteristic symptoms that included headache, fever, joint pains, sweating, loss of appetite, thirst, shivering, and bitter taste in the mouth.

Sources of medicinal plants knowledge and preservations

It was found that majority of indigenous knowledge on anti-malarial plants were restricted to elder members with ages ranging from 51 to 60 years and the younger's have limited contribution in this aspect. The majority of the informants responded that the best means of transferring this traditional medicinal knowledge and skill was at the family level. The information has been orally passed down from family members, particularly grandparents and parents. Nonetheless, some traditional healers kept the knowledge with them. The informants' responses indicated that most of the traditional healers were not interested to transfer their knowledge to interested individuals in the community as they may lose their income or recognition in the community.

As a result of accurate knowledge of the plants and their medicinal properties were held by only a few individuals in the community and without exception of indigenous knowledge which was handed down to some elders throughout generation is at risk of getting lost. On the other hand, informants' report showed that the young generation is not eager to acquire the knowledge and skills of the traditional medicinal plants due to the wrong assumption.

Distribution of medicinal plants in the study area

In the study area, a total of 25 plant species distributed into 22 families were identified. These plant species were used for treating malaria ailment. The family Asteraceae, Euphorbiaceae and Brassicaceae accounted for the highest number of medicinal plants composing 2 species (8%) each and the remaining 19 families composing 76%. The species of Asteraceae family used highly for medicinal purpose may be due to their diversified abundance and adaptation to different habitats in which their continuous harvest do not minimize their abundance

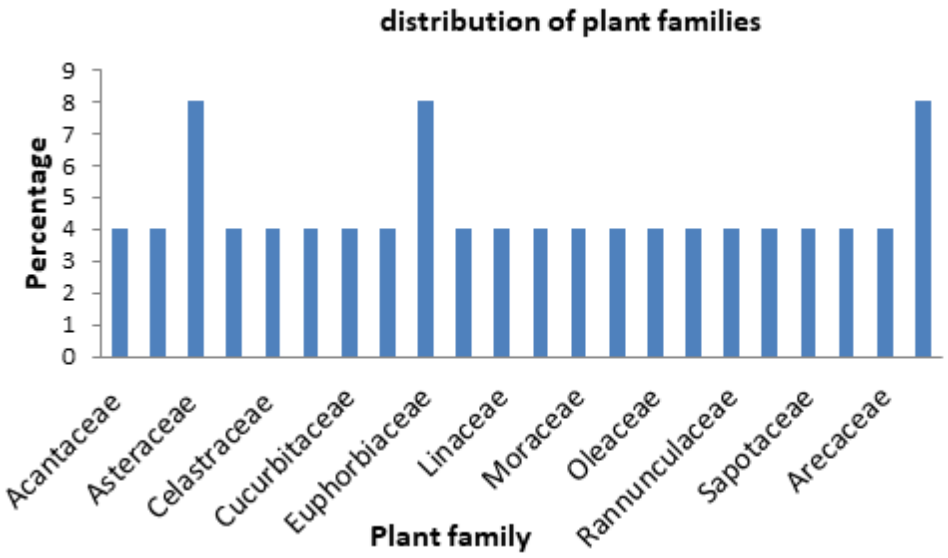

Figure 2. List of families with the species.

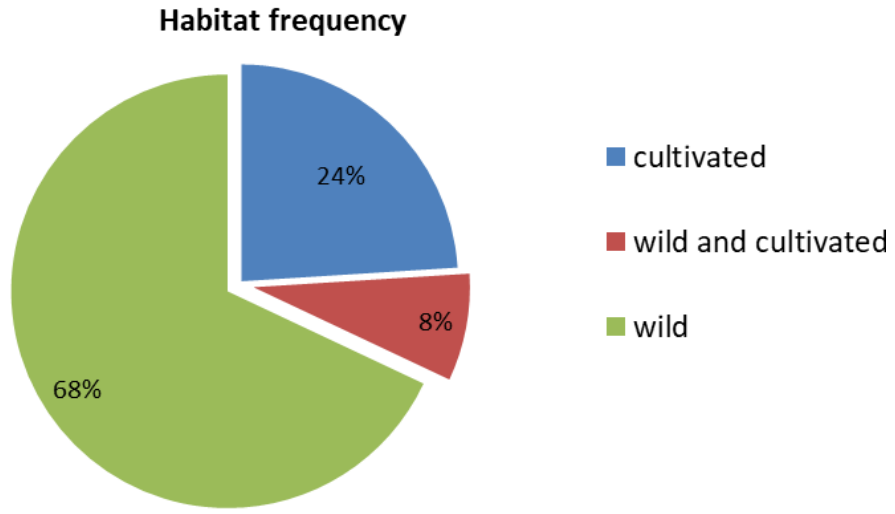

Figure 3. Habitat frequency of medicinal plants.

and most of Asteraceae families are herbs which minimize their destruction for different purposes like trees. The presence of knowledge and practice on medicinal plants by healers shows that the indigenous people of the study area still depend on the traditional medicinal plants (Figure 2).

Habitat variation of medicinal plants

The current finding revealed that most of the medicinal plants obtained from wild 17 (68%) followed by cultivated one 6 (24%). The fact that high number of medicinal plant species was obtained from wilds suggests that the area has good vegetation and wilds are a good option for healers to hide their knowledge from other people which revealed that frequently medicinal plants were collected from the wild in the nearby forest and 68% of the medicinal plants were collected from the natural habitat (Yirga et al., 2011)(Figure 3).

Plant habit

Of the total 25 medicinal plants collected from the study area, 9 (36%) were herb species followed by 7(28%) tree species, 4 (16%) shrubs and 2 (8%) climbers (Figure 4). This shows that most widely used medicinal plants habit in the study area are herbs followed by trees. This may be due to high level of abundance of herb habits in the

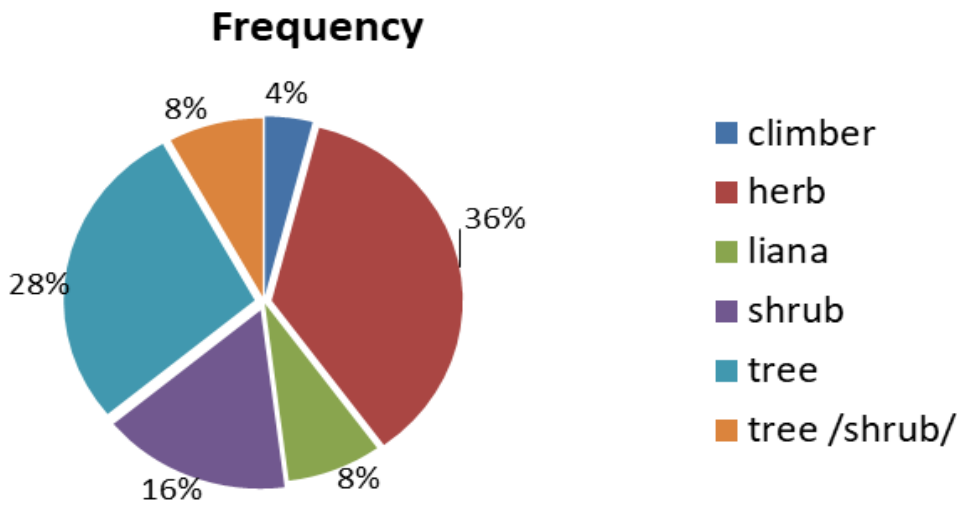

Figure 4. Habits of medicinal plants.

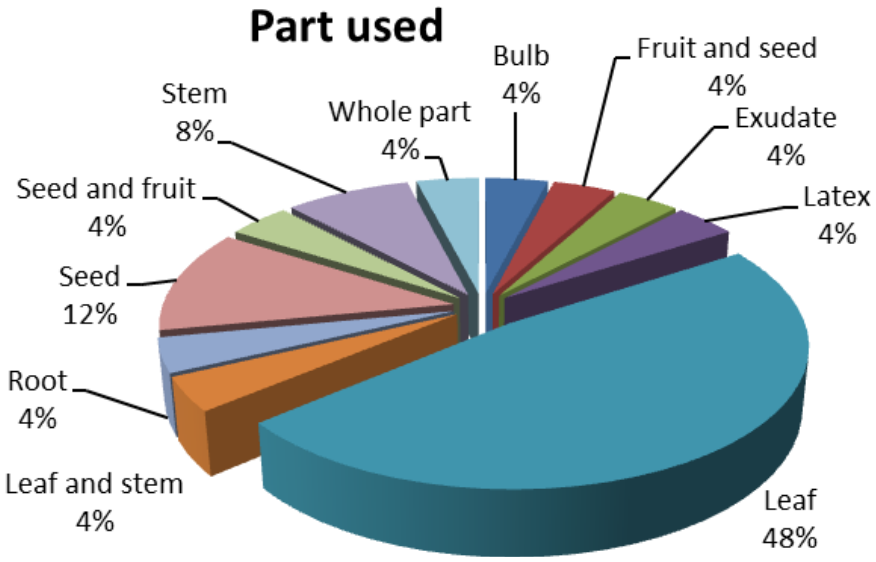

Figure 5. Used parts of medicinal plants.

study area compared to tree and climber species which have less probability of destruction by natural and anthropogenic factors. Relatively high number of herbs and shrubs for medicinal purpose has also been reported previously by Hunde (2004).

Plant parts used

The most widely used part of plant for the preparation of herbal remedies was leaf 48% followed by seed 12%, stem 8% and the remaining plant parts contribute 4% each (Figure 5).

In the study area, leaf is the most cited plant part in medicine preparation which agreed with reports on medicinal plants by Amenu (2007), whereas, studies in Shirka district (Addis et al., 2001) in northern Ethiopia and in Jabitehnan district western Gojjam (Berhanu, 2002). Since herbal preparation that involves roots, rhizomes, bulbs, barks, stems or whole parts have negative effects on the survival of the mother plants. The fear of destruction of medicinal plants due to the nature of plant parts collected for the purpose of medicine seems to be minimal in the area where this study was conducted. Therefore, the traditional medicine practice has little contribution to destruction of the plant species, because collection of leaves has no greater danger to the existence of an individual plant as compared to the

collection of underground parts, stem or whole plant.

Preparation method of medicinal plants

Concerning the preparation of traditional medicine, healers employ various methods of preparation of traditional medicines for malaria treatment. The principal method of traditional medicine preparation reported was squeezing and crushing. This may be due to the possibility of effective extraction of plant ingredients when squeezed or crushed, so that its curative potential would increase.

Preparations may involve using a single plant part or mixtures of different organs of the same plant. In this study, the local people also use some other products as additives in their preparations. For example, water, sugar, salt, milk, and honeys are some of the additives that the local people reported to be used to improve the flavor and reduce adverse effects such as vomiting so that the efficacy of the traditional medicine would be increased. Such additives were also reported by some previous researchers (Abebe, 2001; Giday and Amini, 2003).

Threats to the anti-malarial medicinal plants and indigenous knowledge

In the study area, human and religious factors were found to contribute to the threats that affect medicinal plant species and indigenous knowledge in the study villages. The most serious threat to the existing knowledge and practice on traditional anti-malarial medicinal plants included; agricultural expansion, forest-fire, cultural change, particularly the influence of modernization, accessibility to modern medicine and lack of interests by the younger generations and expansion of protestant religion in which the religious leaders punish healers not to practice were the main problems reported by the informants during the study. Modernization and modification of culture in the area have played a major role in changing the attitude of younger generation to ignore the use of traditional knowledge. Deforestation which is driven by human activities, including agricultural development was reported to be the major threat to the local flora in general and to the anti-malarial medicinal plants in particular.

DISCUSSION

Malaria prevention and treatment

In the study area, traditional medicinal diagnosis is essentially based on systematic interviewing and physical examination if the disease is very serious and the healer thinks that it is the result of evil spirits, magical performances are often carried out to know it clearly. After all, if the healer is unable to ascertain the type of disease, the patient is sometimes referred to the nearby health institution either to bring the result of the diagnosis or to be treated there. The diagnosis of malaria is often simple: intermittent fever and shivering recognized as symptoms of malaria (Dawit Abebe, 1986). The overall conditions of the patient are taken into consideration while measuring out dosages. The major factors that determine whether the treatment is to be given are age, physical fitness, stage of illness, pregnancy and presence or absence of any disease other than the disease to be treated (Dawit Abebe, 1986). For example, drugs that are given through the mouth and the nose are not usually administered to pregnant women unless the patient is in critical situation. Prevention of malaria (as well as other diseases) is commonly practiced by indigenous people using traditional insecticides and insect repellent plants. For example, *A. sativum* is applied on exposed body parts so as to directly attack mosquitoes and other pathogenic insects.

Smoking and growing medicinal plants near the entrances of the fence is also practiced as insect repellents though detailed information is lacking to how it works. The paired comparison of the five species based on their anti-malarial importance only showed that *Cyprus* spp. is most preferred followed by *A. sativum*, *L.sativum* ranked third for prevention and treatment of malaria.

Conservation of medicinal plants

The direct matrix ranking for randomly selected five medicinal plants used to treat malaria on five uses criteria showed that medicinal plants are widely harvested for different purposes. This is particularly true for *E. kebericho* Mesfin., *Croton macrostachyus* Del., *Macaranga capensis* (Baill) Benth. and *Ekebergia capensis* Sparm. Thus, indigenous people use those species for charcoal and fire wood. However, *Cyprus* spp. is extensively used for medicinal purposes by healers only with very little use as fire wood by other people. Generally, the direct matrix ranking shows that those medicinal plants are at conservation risk because of over exploitation and additional uses for different activities.

Conclusions

The number of medicinal plants reported for the prevention and treatment of malaria is a good indicator of the potential that exists locally so long as scientific procedure is added to the indigenous knowledge. In this connection, it is important to develop the indigenous knowledge by focusing on the most popular plants used against malaria including through extraction and

developing phyto medicines. In view of seriousness of malaria in the study area and existence of medicinal plants for the prevention and control of it, research must be enhanced to test activities of those species widely used by the society, namely, *A. sativum, C. macrostachyus, L. sativum, Phytolaca dodecandra, Cyprus* spp. and *E. kebericho* Mesfin. Moreover, a further study on the conservation and sustainable use of medicinal plants is recommended.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Abebe D (2001). The role of medical plants in health care coverage of Ethiopia, the possible integration. In: conservation and sustainable use of medicinal plants in Ethiopia, proceeding of The National work shop on Bio diversity and sustainable use of medicinal plants in Ethiopia, 28 April.

Addis G, Abebe D, Urga K (2001). A survey of traditional medicinal plants in Shirka District, Arsi Zone, Ethiopia, Ethiopian Pharmaceutical Journal 19:30-47.

Afrane A, Little J, Lawson W, Githeko K, Yan G (2008). Deforestation and vectorial vapacity of Anopheles gambiae Giles mosquitoes in Malaria transmission, Kenya. Emerging Infectious Diseases 14:1533-38.

Alemu A, Muluye D, Mihret M, Adugna M, Gebeyaw M (2012) Ten year trend analysis of Malaria prevalence in Kola Diba, North Gondar, Northwest Ethiopia. Parasites and Vectors 5:173.

Amare G (1976). Some common medicinal and poisonous plants used in Ethiopian Folk Medicine (working manual).

Amenu E (2007). Use and Management of Medicinal Plants by indigenous People of EjajiArea (ChelyaWereda) West Shewa, Ethiopia: An Ethnobotanical Approach. M.Sc. Thesis. Addis Ababa, Ethiopia.

Balkew M, Getachew A, Chibsa S, Olana D, Reithinger R, Brogdon W (2012). Insecticide resistance: a challenge to Malaria vector control in Ethiopia. Malaria Journal 11:13.

Berhanu A (2002). Use and conservation of human traditional medicinal plants by indigenous people in JabitehnaWoreda, West Gojjam: with special emphasis to plant species that are used as anti-Malarial treatments, insecticides and insect repellents, (M.Sc thesis, unpublished).

Central Statistical Agency (CSA) (2016). The Ethiopian Demographic and Health Survey, Central Statistical Agency, Addis Ababa, Ethiopia.

Dawit A (1986). Traditional medicine in Ethiopia: the attempts being made to promote it for effective and better utilization. SINET: Ethiopia. Journal Science 9:61-69.

Deressa W, Ali A, Berhane Y (2006). Review of the interplay between population dynamic and Malaria transmission in Ethiopia. Ethiopia Journal Health Development 20:137-144.

Geoffrey C, Kirby G (1996). Medicinal plant and the control of protozoa disease with particular reference to Malaria. Transactions of the Royal Society of Tropical Medicine and Hygiene 90:605-609.

Giday M, Amini G (2003). An ethno botanical survey on plants of veterinary importance in two weredas of Southern Tigray, Northern Ethiopia. SINET: Ethiopia Journal Science 26(2):123-136.

Hostettman K, Marston A (2002). Twenty years of research into medicinal plants: resultsand perspectives. Phytochemistry Reviews 1:275-285.

Hunde D (2004). Use and management of Ethno veterinary medicinal plants of indigenous people in Boosat, welenchiti, Ethiopia. Journal. Biological Science 3(2):11-132.

Karou D, Nadembega W, Ouattara L, Ilboudo P, Canini A, Nikiéma J, Simpore J, Colizzi V, Traore A (2007). African ethnopharmacology and new drug discovery. Medicinal Plant Science and Biotechnology 1:61-69.

Massebo F, Balkew M, Gebre-Michael T, Lindtjorn B (2013). Blood meal origins and insecticide susceptibility of Anopheles arabiensis from Chano in South-West Ethiopia. Parasites and Vectors 6:44.

Ministry Health State of Federal Democratic Republic of Ethiopia (MOH) (2012). National Malaria guidelines.3rd edition. Addis Ababa: Commercial Printing enterprise; January, 2012.

Ngutaa J, Mbariaa J, Gakuyab D, Gathumbic P, Kiamad S (2010). Anti-Malarial herbal remedies of Msambweni, Kenya. Journal of Ethnopharmacology 128:424-432.

Otten M, Aregawi M, Were W, Karema C, Medin A, Bekele W, Jima D, Gausi K, Komatsu R, Korenromp E, Low-Beer D, Grabowsky M (2009). Initial evidence of reduction of Malaria cases and deaths in Rwanda and Ethiopia due to rapid scale-up of Malaria prevention and treatment Malaria Journal 8:14.

Phillipson J, Wright C (1991). Can ethno pharmacology contribute to the development of anti-Malarial agents? Journal of Ethnopharmacology 32:155-165.

Tamene B (2000). A Floristic Analysis and Ethno botanical Study of the Semi- Wet land of Cheffa Area, South Wello, Ethiopia.M. Sc. Thesis, Addis Ababa University.

World Health Organization (WHO) (2015). World Malaria Report, Geneva, Switzerland, 2015. http://www.who.int/ world-malaria-report-2015.

World Health Organization (WHO), (2013). World Malaria Report 2013. ⟨http://apps.who. int/iris/bitstream/10665/97008/1/9789241564694_eng.pdf⟩ (accessed on 3.04.18.

Yirga G, Mekonen T, Mezgebe K (2011). Survey of medicinal plants used to treat human ailments in Hawzen district, Northern Ethiopia. International Journal of Biodiversity and Conservation 3(13):709-714.

Appendix 1. The list of medicinal plants recorded in the study area for malaria prevention and treatment.

Local name	Scientific Name	Family	Habit	Habitat	Parts used	Route
Nech Shinkurit	*Allium sativum L.*	Alliaceae	H	Cu	Bulb	Oral
	Bothriocline schimperi Olivo & Hiern Ex. Benth.	Asteraceae	H	Wi	Leaf	Oral
Lol	*Ekebergia capensis Sparm*	Meliaceae	T	Wi	Leaf	Oral
	Pouteria adolfi-friederici (Engl.) Baehni.	Sapotaceae	T	Wi	Leaf	Oral
Yewef enqur	*Commelina latifolia Hochst. Ex A. Rich.*	Commelinaceae	H	Wi	Leaf and stem	Oral
Sinafich	*Brassica nigra (L.) Koch In Rohling.*	Brassicaceae	H	Cu	Fruit Seed	Oral
Papaya	*Carica papaya L.*	Carricaceae	T	Cu	Leaf	Oral
Nech yeazo areg	*Clematis longicaudata Steud. ex A. Rich*	Rannunculaceae	liana	wi	Leaf	Oral
Misirich	*Clerodendrum myricoides (Hochst.) R. Br*	Lamiaceae	Sh	wi	Leaf	Oral
Bissana	*Croton macrostachyus Del.*	Euphorbiaceae	T	Wi	Leaf	Oral
	Cyperus spp.	Cyperaceae	H	wc	Whole part	Nasal
Kebericho	*Echinops kebericho Mesfin.*	Asteraceae	H	Wi	Root	Oral
Shola	*Ficus* spp.	Moraceae	T	Wi	Latex	Oral
Feto	*Lepidium sativum L.*	Brassicaceae	H	cu	Seed	Oral
Teliba	*Linum usitatissimum L.*	Linaceae	H	Cu	Seed	Oral
	Macaranga capensis (Baill) Benth	Euphorbiaceae	T	wi	Exudate	Oral
Kelewa	*Maesa lanceolata Forssk.*	Myrsinaceae	T/sh	Wi	Leaf	Oral
Atat	*Maytenus gracilipes (Welw. Ex Oliv.) Exell.*	Celastraceae	T/Sh	Wi	Leaf	Oral
Areg resa	*Momordica foetida*	Cucurbitaceae	Cl	Wi	Seed, fruit	Nasal
Timbaho	*Nicotiana tabacum*	Solanaceae	H	Cu	Leaf	Nasal
Zembaba	*Phoenix reclinata*	Arecaceae	T	wi	Stem	Oral
Kitikta	*Dodonea angustifolia L*	Sapindaceae	Sh	wi	Leaf	Oral
Tembelel	*Jasminum abyssinicum Hochst*	Oleaceae	Liana	wi	Leaf	Oral
Simiza	*Justicia schimperiana (Hochst. ex Nees*	Acanthaceae	Sh	wi	Leaf	Oral
Endod	*Phytolaca dodecandra L'Herit*	phytolacaceae	Sh	wc	Seed	Oral

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