TANG HUMANITAS MEDICINE

Review

Tinjute [Labiatae; (*Otostegia integrifolia*)]: A versatile Ethiopian ethnomedicinal plant – a systematic review of the scientific evidences

Kaliyaperumal Karunamoorthi

Unit of Medical Entomology and Vector Control, Department of Environmental Health Science& Technology, College of Public Health and Medical Sciences, Jimma University, Jimma, Ethiopia

ABSTRACT

Despite the remarkable advancements of the modern medicine, the traditional system of medicine (TSM) still serves as a potential primary health care modality in the in low- and middle-income countries. The recent reports suggest that there is a renewed interest has been observed towards TSM in the developed countries too, because of the adverse side-effect of modern medicines. Medicinal plants have been widely serving as a rich source of therapeutic agent. Ethiopia is one of the most reserves rich countries in the world. It is renowned for well-diversified and natural resources in terms of its unique flora and fauna. Ethiopian deep-rooted tradition and culture largely depends on the usage of plants for their religious ceremonies, impressive festivals, traditional medicinal uses and other basic necessities. The present scrutiny is an attempt to understand the omnipotent nature of an Ethiopian Ethnomedicinal plant called Tinjute [vernacular name (local native language, Amharic); *Otostegia integrifolia*]. There are several studies suggest that Tinjute can be used as a natural medicine or health-promoting agents for various disorders and ailments. Nevertheless, in Ethiopia, it is renowned as an insect repellent to drive-away insect vector of diseases, particularly mosquitoes in the early evening. However, there are many more issues and challenges which must be urgently addressed to scientifically formulate various potent, efficacious, safe and highly selective phytotherapeutic agents and insects' repellent from the Tinjute plant in the near future.

Keywords traditional system of medicine, *Otostegia integrifolia*, Tinjute, insect repellent, Ethiopia

INTRODUCTION

Since ages, humans have relied on nature for their basic amenities like comestibles, shelters, clothing, means of transportation, fertilizers, flavours, and fragrances, and not the least, medicines (Gurib-Fakim, 2006). Medicinal plants have formed the basis of sophisticated traditional medicinal practices, which have been used for thousands of years by people in China, India, Africa, and the rest of the world (Sneader, 2005). Early humans acquired the knowledge on plants' therapeutic values through many years of vigilant observations, experience and trial and error experiments (Karunamoorthi et al., 2013a, 2012; Martin, 1995; Sofowora, 1982). Plant kingdom remains to be a repository of modern medicine due to their rich resource of bio-active molecules and secondary metabolites. Since time immemorial, each and every society has had their unique way of indigenous health practice system by using plants as a curative source to treat various ailments. However, the Chinese, Indian, and African traditional systems of medicine are globally renowned (Karunamoorthi et al., 2012).

Ethiopia: A cradle of medicinal plants

The Ethiopian highlands, which are the most extensive of the African mountain region, are presumed to be covered by mountain forests. Historically, they have been coined as the water tower of east and northeast Africa - a notion, which refers to the fact that a number of rivers have their sources in the highlands. The Afromontane forests of Ethiopia, render ecological, socioeconomic and medicinal significance for the humankind. Ethiopia is rich in biodiversity with high endemism. The richness in flora and fauna is a reflection of the diverse ecological setting, climate and topography found in the country. The Ethiopian flora is estimated to contain 6500 to 7000 species of higher plants, of which about 12 percent are endemic. The country has the fifth largest flora in tropical Africa (Mekonnen and Hailemariam, 2000). Plants have been used as a source of medicine in Ethiopia from time immemorial to treat various ailments (Karunamoorthi and Tsehaye, 2012).

Tinjute: An Ethiopian medicinal plant

Genus *Otostegia* belongs to the family *Lamiaceae*, which consists of about 180 genera and over 3500 species. The *Otostegia* comprises about twenty species, among them fifteen are endemic to the northern part of tropical Africa and Southwestern and Central Asia, while the remaining five species have been reported in the flora of Ethiopia. Naturally, the species of *Otostegia* are medicinally decisive and, have been traditionally been used as an ophthalmia, mosquito repellency, antimicrobial, antihyperglyceamic, and antioxidant activity to prevent different kinds of sickness and disorders (Khan and Syed, 2013). Besides, its chemical constituents have also shown to possess antiulcer, antispasmodic, antidepressant, anxiolytic and sedative activities (Vural et al., 1996). In Ethiopia, *O. integrifolia* is commonly known by its vernacular

*Correspondence: Kaliyaperumal Karunamoorthi
E-mail: karunamoorthi@gmail.com
Received December 4, 2013; **Accepted** May 2, 2014; **Published** May 31, 2014
doi: http://dx.doi.org/10.5667/tang.2013.0033

Table 1. Scientific classification of Tinjute

Kingdom	Plantae
Class	Magnoliopside
Order	Lamiales
Family	Lamiaceae
Genus	Otostegia
Species	Integrifolia - Benth

name 'Tinjute' (Amharic - local language). It is an herbaceous plant that grows in the wild, but it is also cultivated in the gardens. The usage of plants for various day-to-day activities/ceremonies is deep-rooted in the Ethiopian tradition and culture and they use numerous plants as herbs to treat various sicknesses and disorders. Among them *O. integrifolia* is well-known for its pleasant odour, omnipotent medicinal values and insecticidal properties.

Taxonomy

O. integrifolia is a synonym of *Rydingia integrifolia* (Benth.) Scheen and V.A. Albert. It is easily recognized and stands out due to the grayish color of the leaves. The yellow flowers are also showy when in full bloom (Getahun, 1976). It is one of the plants very often administered in the Ethiopian traditional system of medicine (Table 1).

Chemical constituents of Tinjute

The essential oil and chloroform extract of air-dried leaves of *O. integrifolia* were investigated using analytical and preparative gas chromatography (GC), GC-mass spectrometry and Nuclear Magnetic Resonance techniques. A total of 40 constituents, including monoterpenes, sesquiterpenes, diterpenes and their derivatives were identified (Tesso and König, 2004). However, the five of the chemical compounds viz. axinyssene, otostegindiol, pretostegindiol, pentatriacontane and stigmasterol are observed to be the potential sources of pharmaceutics and medicine, to treat various illnesses (Fig. 1).

As an insect repellent plant

Over decades, the vector-borne diseases impose a serious public health concern to human beings in terms of considerable morbidity and mortality. The concomitant effects of global warming linked with climate change has further fueled the emergence and resurgence of several insect-borne diseases like malaria, plague, filariasis, trypanosomiasis, leishmaniasis and many arbo-viral diseases particularly dengue fever, yellow fever and chikungunya fever (Karunamoorthi, 2013). They account for nearly 17% of the global burden of infectious diseases (WHO, 2006). Besides, they also impose severe negative socioeconomic impact in the endemic regions (Karunamoorthi and Sabesan, 2009). Therefore, it is mandatory to minimize the global burden of these diseases by applying appropriate integrated vector control strategies and personal protection interventions.

An insect repellent is a substance applied to skin, clothing, or other surfaces to keep away insects from landing or climbing on that surface. The primary aim of the repellent usage is to reduce the man-vector contact and eventually to minimize the considerable annoying biting nuisance and disease transmission (Karunamoorthi and Sabesan, 2009). Repellents can be neither chemical nor natural (plant-based) and they are very supportive whenever and wherever other sorts of personal protection interventions are ineffective, unfeasible and impracticable (Karunamoorthi and Sabesan, 2010; Karunamoorthi et al., 2014). It is important to note that the existing synthetic repellents are usually inaccessible as well as unaffordable. Besides, they also pose a high level of residual toxicity and adverse side-effect (Karunamoorthi, 2011) for infants and elderly people and a few of them needed electricity also for their usage (Karunamoorthi et al., 2008a).

Since ancient time plants have been used to repel or kill the blood-sucking insects (Karunamoorthi et al., 2008b). Plant-based repellent products are easily available, locally known, and culturally acceptable (Karunamoorthi et al., 2010a). In Ethiopia, the rural population has often been using these plant-based materials as repellents by means of smoldering the dried plant parts (leaves, stem, root, bark and resin) on the traditional charcoal stove in the early evening to minimize the man-vector contact (Fig. 2). It is interesting to note that these low-cost repellant materials are abundantly obtainable in almost all the Ethiopian markets (Fig. 3). Ethnobotanical surveys in Ethiopia (Karunamoorthi et al., 2009a, b; Kidane et al., 2013; Teklay et al., 2013) and Eretria (Waka et al., 2004) found that the local

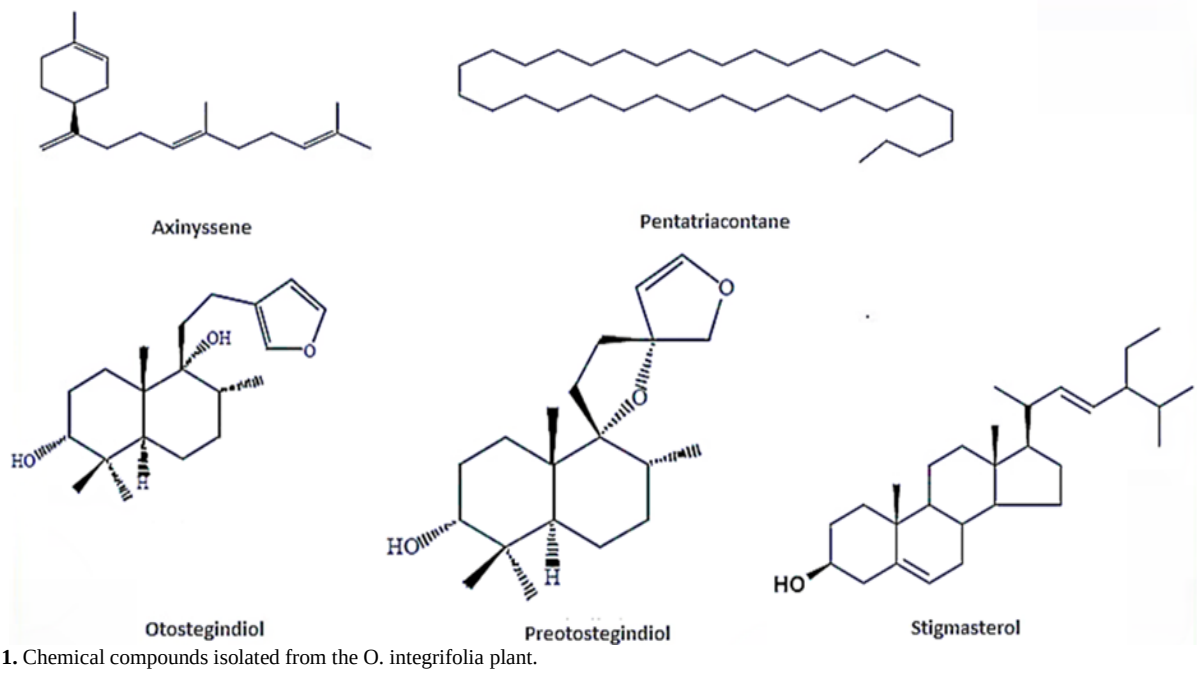

Fig. 1. Chemical compounds isolated from the O. integrifolia plant.

Fig. 2. An Ethiopian women describing about the traditional usage of repellent plants by means of smoldering/burning on the traditional charcoal stove.

residents have been using *O. integrifolia* as a potential traditional medicine to alleviate various illnesses and disorders (Kidane et al., 2013; Teklay et al., 2013) and insect repellent (Karunamoorthi et al., 2009a, b) to drive-away insects particularly mosquitoes by burning the dried plant parts as well as by hanging the plant around the main doors, windows and nearby sleeping cots in the early evening hours in Eretria (Waka et al., 2004).

In Ethiopia, an experiment was conducted by applying the smoke into the repellent "test" mosquito cage by direct burning of 25gm of dried leaves of *O. integrifolia* until plant materials are completely burnt. The result clearly revealed that the leaves of *O. integrifolia* have a potent repellent efficiency against *Anopheles arabiensis*, a principal malarial vector in Ethiopia (90.10%) and was the most effective (Karunamoorthi et al., 2008a). Since people have been using *O. integrifolia* for some medicinal purposes, no side effects have been reported. Therefore, it is suggested that this plant can be used as a safer alternative to modern synthetic chemical repellents against insect vector of diseases (Karunamoorthi et al., 2010).

The *O. integrifolia* has insecticidal properties and is often used as fumigant for pots and houses (Karunamoorthi et al., 2009a, b). Indeed the repellent activity of *O. integrifolia* was scientifically proved against *An. arabiensis*, however, further research/studies are warranted to examine, isolate, characterize and identify the untapped pool of bio-active molecules, which are accountable for the repellency effect. Furthermore, conducting more laboratory and field based studies are inevitable for human safety concerns (Karunamoorthi and Husen, 2012).These kinds of pioneer research could pave the way to formulate a new class of potent insect' repellent from *O. integrifolia* in the near future (Karunamoorthi, 2012).

As a traditional phytotherapeutic agent

Over centuries, cultures around the world have learnt the usage of medicinal plants to fight against illness and to maintain good health. In the resource-limited settings, these plants are one of the longest-serving companions for the rural poor due to their easier accessibility and affordability (Karunamoorthi et al., 2013; Roberson, 2008). Despite the recent scientific advancement and globalization, the system of traditional medicine and complementary/alternative medicine is considered to be a primary health care modality in the resource-constrained health care settings (Roberson, 2008). The WHO estimates that, even now more than 80% of the world's population relies on traditional healing modalities and herbals for primary health care and wellness (WHO, 2002). The heavy reliance on medicinal plants is partly owing to the high cost of modern drugs, limited or inaccessibility of modern healthcare facilities (Cunningham, 1993; WHO, 2003).

In the past, the importance of traditional medicinal plants and their phytotherapeutic values have often been disregarded and undervalued. However, in the recent years the revitalization and renewed interest on medicinal plants has been well-observed among the health conscious public and scientific community (Karunamoorthi et al., 2012), due to the long-term adverse consequences of modern chemotherapeutic agents. In Ethiopia, 70% and 90% of human and livestock population depend on the traditional system of medicine, respectively (Bekele, 2007).

Antimalarial activity

Malaria continues to be a major global public health problem and about 3.3 billion people are at the risk in 104 endemic countries. It remains to be one of the important causes of maternal and childhood morbidity and mortality in sub-Saharan Africa and particularly Ethiopia (Karunamoorthi et al., 2010b).It is a disease of poverty and a cause of poverty which inflicts a serious negative impact on global public health and it has also hampered socioeconomic development among the poorest countries of the world (Karunamoorthi, 2012b).Globally, over thousand plants have been used as potential antimalarials in the resource-poor settings due to fragile/limited health-care systems, which quite often fail to meet the expectations of the needy poor people (Karunamoorthi et al., 2013b).

At the moment, there is a growing acceptance and keen interest towards traditional system medicines and it has led many researchers to concentrate on traditional antimalarial phytotherapy. Furthermore, the existing potent antimalarial artemisinin combination therapy is critically short supply unaffordable and inaccessible too. The recent reports suggest the emergence and spread of *Plasmodium falciparum* and *Plasmodium vivax* multidrug resistance malaria on Thailand-Myanmar and Thailand-Cambodia borders, the hotspot of multidrug resistance parasites (Bhumiratana et al., 2013; Karunamoorthi and Husen, 2012). Therefore, identifying the affordable and easily accessible potent antimalarials is a matter of grave concern. The analysis of traditional medicines that are employed for the treatment of malaria represents the potentiality for the discovery of lead molecules and henceforth for further development of potential antimalarial drugs (Muthaura et al., 2007). The new antimalarials are vital to combat and contain the emergence, resurgence and spread of multi-drug resistance strains in the near future.

In Ethiopian traditional medicine, the leaves of *O. integrifolia* are used for the treatment of several diseases including malaria. In an ongoing search for effective, safe and cheap antimalarial agents from plants, the 80% methanol leaf extract *O. integrifolia* was tested for its in vivo antimalarial activity against *Plasmodium berghei*. The results are encouraging and the extract exhibited potent antiplasmodial activity resulted in the isolation of a labdane diterpenoid identified as otostegindiol. Conclusively, the reputed antimalarial effect of *O. integrifolia* may be attributed in whole or in part to the presence of the diterpenoid otostegindiol.

The genuine antiplasmodial activity along with its safety profile observed in the study could make the leaf extract of *O. integrifolia* a potential addition to the antimalarial armamentarium, and shall also provide a scientific support for the ethnomedicinal use of the plant (Endale et al., 2013). Besides, otostegindiol can be useful as a chemical and/or

Fig. 3. Ethiopian vendors selling their repellant plants in the market of the Jimma town, Ethiopia .

biological marker to guarantee the quality of the herbal product. Therefore, *O. integrifolia* may serve as potential candidates for the development of efficacious as well as safe phytomedicines (Endale et al., 2013).

Though many believe that the usages of medicinal plants are relatively safe, which have folklore reputations for antimalarial properties, many herbs may also be potentially toxic due to their intrinsic adverse side-effects. Therefore herbal-derived remedies need a powerful and deep assessment of their pharmacological qualities in order to establish their mode of action, safety, quality and efficacy. World-wide, over thousand plant species have been used as a source of traditional antimalarial phytotherapy for many centuries and in the future, plant-derived compounds will still be an essential aspect of the therapeutic array of antimalarials. However, their clinical efficacy, quality and safety need to be scientifically evaluated and proved (Karunamoorthi et al., 2013b).

Other medicinal properties of Tinjute

Research has indicated that one of the chemical constituents of *O. integrifolia* called stigmasterol may be useful in prevention of certain cancers, including ovarian, prostate, breast, and colon cancers. Studies with laboratory animals fed stigmasterol found that both cholesterol and sitosterol absorption decreased 23% and 30%, respectively. It was demonstrated that it inhibits several pro-inflammatory and matrix degradation mediators typically involved in osteoarthritis-induced cartilage degradation (Gabay et al., 2010). It also possesses potent antioxidant, hypoglycemic and thyroid inhibiting properties (Panda et al., 2009). Stigmasterol is the precursor of anabolic steroid boldenone and oldenone undecylenate is commonly used in veterinary medicine to induce growth in cattle, but it is also one of the most commonly abused anabolic steroids in sports (Draisci et al., 2007; Gallina et al., 2007; Ros et al., 2007). In Saudi Arabia, the irritated eyes of cows, goats, and sheep can be rinsed with leaf infusion of the *O. integrifolia* (Abulafatih, 1987). The roots of *O. integrifolia* are used for treating lung diseases too.

Miscellaneous purposes in Ethiopia

Recent investigations indicate that the leaves of the *O. integrifolia* was used for the treatment of tonsillitis, uvulitis, lung diseases, stomachache, and hypertension (Tesso and König, 2004; Giday et al., 2007; Teklehaymanot et al., 2007; Andemariam, 2010). In Northern Ethiopia, where it is common, it is used to smoke utensils to flavor them and to sterilize them at the same time. According to the local understanding, the practice of smoking the vessel by burning wooden chips of specific trees and shrubs has an advantage of imparting special taste and odour to the product, and to disinfect the vessels, thus reducing the numbers of micro-organisms and thereby extending the shelf life of the product (Alemayehu, 2009). Clothes are also smoked; perhaps the most common practice is the ritual custom that a mother is smoked with “Tinjute” on the tenth day after giving birth to a child. She is "cleansed" with the smoke after that process she can leave her confinement and again resume a normal daily life (Getahun, 1976; Wilson and Mariam, 1979).

CONCLUSION

Though utilization of medicinal plants is not a new phenomenon, now there is more consciousness among the people about the adverse effects of synthetic products as well user-friendly nature of plant-based products than ever before. In Ethiopia, *O. integrifolia* is one of the reputable plants for its phytotherapeutic potentialities and insecticidal properties. However, there are only a limited number of studies have been reported to authenticate the omnipotent properties of *O. integrifolia*, particularly, its traditional usage as antimalarial, disinfectant and insecticidal agents. Therefore, further studies are warranted by using the existing modern scientific methods and innovative techniques to prove the potency, safety and feasibility in the future.

ACKNOWLEDGEMENTS

I would like to thank Mrs. Melita Prakash for her sincere assistance in editing the manuscript. I sincerely would like to acknowledge my friend Mr. Embialle Mengiste who has shared his knowledge and experience on the *O. integrifolia* plant. Without his contributions, this editorial would have been impossible. My last but not the least heartfelt thanks go to my colleagues of our Department of Environmental Health Science, College Public Health and Medicine, Jimma University, Jimma, Ethiopia, for their kind support and cooperation.

CONFLICT OF INTEREST

The authors have no conflicting financial interests.

REFERENCES

Abulafatih HA. Medicinal Plants in Southwestern Saudi Arabia. Econ Botany. 1987;41:354-360.

Alemayehu YA, Wurzinger M, Tegegne A, Zollitsch W. Handling, processing and marketing of milk in the north western Ethiopian highlands. Livestock Research for Rural Dev. 2009;21:97.

Andemariam SW. Legislative regulation of traditional medicinal knowledge in Eritrea vis-à-vis Eritrea's commitments under the convention on biological diversity: Issues and alternatives. Law Environ Dev J. 2010;6:130-162.

Bekele E. Study on actual situation of medicinal plants in Ethiopia. Japan Association for International Collaboration of Agriculture and Forestry. 2007. Available at: http://www.endashaw.com (accessed on 29th January 2011).

Bhumiratana A, Intarapuk A, Sorosjinda-Nunthawarasilp P, Maneekan P, Koyadun S. Border malaria associated with multidrug resistance on Thailand-Myanmar and Thailand-Cambodia borders: transmission dynamic, vulnerability, and surveillance. Biomed Res Int. 2013;2013:ID363417.

Cunningham AB. African Medicinal Plants: Setting Priorities at the Interface between Conservation and Primary Health Care. UNESCO. 1993.

Draisci R, Merlanti R, Ferretti G, Fantozzi L, Ferranti C, Capolongo F, et al. Excretion profile of boldenone in urine of veal calves fed two different milk replacers. Analytica Chimica Acta. 2007;586:171-176.

Endale A, Bisrat D, Animut A, Bucar F, Asres K. In vivo Antimalarial Activity of a Labdane Diterpenoid from the Leaves of Otostegia integrifolia Benth. Phytother Res. 2013;27:1805-1809.

Gabay O, Sanchez C, Salvat C, Chevy F, Breton M, Nourissat G, et al. Stigmasterol: a phytosterol with potential anti-osteoarthritic properties. Am J Clin Nutr. 2010;18:106-116.

Gallina G, Ferretti g, Merlanti R, Civitareale C, Capolongo F, Draisci R, Montesissa C, Boldenone, Boldione, and Milk Replacers in the Diet of Veal Calves: The Effects of Phytosterol Content on the Urinary Excretion of Boldenone Metabolites. J Agric Food Chem. 2007;55:8275-8283.

Getahun A. Some common medicinal and poisonous plants used in Ethiopian folk medicine. (Addis Ababa, Ethiopia: Addis Ababa University), 1976.

Giday M, Teklehaymanot T, Animut A, Mekonnen Y. Medicinal plants of the Shinasha, Agew-awi and Amhara peoples in North West Ethiopia. J Ethnopharmacol. 2007;110:516-525.

Gurib-Fakim A. Medicinal plants: Traditions of yesterday and drugs of tomorrow. Mol Aspects Med. 2006;27:1-93.

Karunamoorthi K, Deboch B, Tafere Y. Knowledge and practice concerning malaria, insecticide-treated net (ITN) utilization and antimalarial treatment among pregnant women attending specialist antenatal clinics. J Pub Health. 2010;18:559-566.

Karunamoorthi K, Girmay A, Fekadu S. Mosquito Repellent Activity of Essential oil of Ethiopian Ethnomedicinal Plant against Afro-tropical Malaria Vector *Anopheles arabiensis*. J King Saud University - Sci. 2014. [Article In Press]

Karunamoorthi K, Husen E. Knowledge and Self-reported Practice of the Local Inhabitants on Traditional Insect Repellent Plants in Western Hararghe Zone, Ethiopia. J Ethnopharmacol. 2012;141:212-219.

Karunamoorthi K, Ilango K, Endale A. Ethnobotanical survey of knowledge and usage custom of traditional insect/mosquito repellent plants among the Ethiopian Oromo ethnic group. J Ethnopharmacol. 2009;125:224-229.

Karunamoorthi K, Ilango K, Murugan K. Laboratory evaluation of traditionally used plant-based insect repellent against the malaria vector *Anopheles arabiensis* Patton (Diptera: Culicidae). Parasitol Res. 2010;106:1217-1223.

Karunamoorthi K, Jegajeevanram K, Jerome X, Vijayalakshmi J, Melita L. Tamil Traditional Medicinal System – Siddha: An Indigenous Health Practice in the International Perspectives. TANG. 2012;2:e12.

Karunamoorthi K, Jegajeevanram K, Vijayalakshmi J, Embialle M. Traditional Medicinal Plants: A Source of Phytotherapeutic Modality in the Resource-Constrained Health Care Settings. J Evidence-Based Complemen Altern Med. 2013;18:67-74.

Karunamoorthi K, Mulelam A, Wassie F. Assessment of knowledge and usage custom of traditional insect/mosquito repellent plants in Addis Zemen Town, South Gonder, North Western Ethiopia. J Ethnopharmacol. 2009;121:49-53.

Karunamoorthi K, Mulelam A, Wassie F. Laboratory evaluation of traditional insect/mosquito repellent plants against *Anopheles arabiensis*, the predominant malaria vector in Ethiopia. Parasitol Res. 2008;103:529-534.

Karunamoorthi K, Ramanujam S, Rathinasamy R. Evaluation of leaf extracts of *Vitex negundo* L. (Family: Verbenaceae) against larvae of *Culex tritaeniorhynchus* (Insecta; Diptera; Culicidae) and repellent activity on adult vector mosquitoes. Parasitol Res. 2008;103:545-550.

Karunamoorthi K, Sabesan S, Jegajeevanram K, Vijayalakshmi J. The Role of Traditional Anti-malarial Plants in the Battle against Global Malaria Burden. Vector Borne Zoonotic Dis. 2013;13:521-544.

Karunamoorthi K, Sabesan S. Field trials on the efficacy of DEET impregnated anklets, wristbands, shoulder, and pocket strips against mosquito vectors of disease. Parasitol Res. 2009;105:641-645.

Karunamoorthi K, Sabesan S. Laboratory evaluation of Dimethyl phthalate treated wristbands against three predominant mosquito (Diptera: Culicidae) vectors of disease. Eur Rev Med Pharmacol Sci. 2010;14:443-448.

Karunamoorthi K, Tsehaye E. Ethnomedicinal Knowledge, Belief and Self-reported Practice of local inhabitants on Traditional Antimalarial Plants and Phytotherapy. J

Ethnopharmacol. 2012;141:143-150.

Karunamoorthi K. Global Malaria Burden: Socialomics Implications. J Socialomics. 2012;1:e108.

Karunamoorthi K. Plant-Based Insect Repellents: Is That a Sustainable Option to Curb the Malaria Burden in Africa? J Med Aromat Plants. 2012b;1:e106

Karunamoorthi K. Yellow Fever Encephalitis: An Emerging and Resurging Global Public Health Threat in a Changing Environment. In Encephalitis, Sergey Tkachev ed. (Croatia: licensee InTech), pp. 207-230, 2013.

Karunamoorthi K.Vector Control: A Cornerstone in the Malaria Elimination Campaign. Clin Microbiol Infect. 2011;17:1608-1616.

Khan F, Syed F. Bioactive Constituents from Genus Otostegia. SARJ of Physical Sci. 2013;1:15-25.

Kidane D, Tomass Z, Dejene T. Community knowledge of traditional mosquito repellent plants in Kolla Temben District, Tigray, Northern Ethiopia. Sci Res Essays. 2013;8:1139-1144.

Martin GJ. Ethnobotany: A method Manual. (London, UK: Chapman and Hall), 1995.

Mekonnen A and Hailemariam K. Baseline and Climate Change Scenarios for Ethiopia. (Addis Ababa, Ethiopia: National Meteorological Services Agency), 2000.

Muthaura CN, Rukunga GM, Chhabra SC, Omar SA, Guantai AN, Gathirwa JW, Tolo FM, et al. Antimalarial Activity of Some Plants Traditionally used in Meru district of Kenya. Phytother Res. 2007;21:860-867.

Panda S, Jafri M, Kar A, Meheta BK. Thyroid inhibitory, antiperoxidative and hypoglycemic effects of stigmasterol isolated from Butea monosperma. Fitoterapia. 2009;80:123-126.

Roberson E. Medicinal Plants at Risk. Nature's Pharmacy, Our Treasure Chest: Why We Must Conserve Our Natural Heritage. (Tucson ,USA: Center for Biological Diversity), 2008.

Ros MM, Sterk SS, Verhagen H, Stalenhoef AF, de Jong N. Phytosterol consumption and the anabolic steroid boldenone in humans: a hypothesis piloted. Food Addit Contam. 2007;24:679-684.

Sneader W. Drug Discovery: a History. (Chichester, UK: Wiley), 2005.

Sofowora A. Medicinal plants and Traditional medicine in Africa. (New York, USA: Wiley), pp 255-256, 1982.

Teklay A, Abera B, Giday M. An ethnobotanical study of medicinal plants used in Kilte Awulaelo District, Tigray Region of Ethiopia. J Ethnobiol Ethnomed. 2013;9:65.

Teklehaymanot T, Giday M, Medhin G, Mekonnen Y. Knowledge and use of medicinal plants by people around Debre Libanos monastery in Ethiopia. J Ethnopharmacol. 2007;111:271-283.

Tesso H, König WA. Terpenes from Otostegia integrifolia. Phytochem. 2004;65:2057-2062.

Vural K, Ezer N, Erol K, Sahin FP. Anxiolytic and antidepressant activity of some Ballota species. J Fac Pharm Gazi. 1996;13:29-32.

Waka M, Hopkins RJ, Curtis C. Ethnobotanical survey and testing of plants traditionally used against hematophagous insects in Eritrea. J Ethnopharmacol. 2004;95:95-101.

WHO. Pesticides and their application. (Geneva, Switzerland: World Health Organization), 2006.

WHO. WHO Traditional Medicine Strategy 2002-2005. (Geneva, Switzerland: World Health Organization), 2002.

WHO. Traditional Medicine: Fact Sheet No 134. 2003. Available at: http://www.who.int/mediacentre/factsheets/2003/fs134/en/ (accessed on 01th November 2014).

Wilson RT, Mariam WG. Medicine and Magic in Central Tigre: A Contribution to the Ethnobotany of the Ethiopian Plateau. Econ Botany. 1979;33:29-34.