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INSIGHT TO LEMONGRASS ESSENTIAL OIL AS PHYTOMEDICINE: STATE OF THE ART AND FUTURE PERSPECTIVES

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ABSTRACT

Lemongrass oil (LGO) is extracted from fresh leaves of *Cymbopogon citratus* by steam distillation. Chemical constituents of Lemongrass oil are myrcene, citronellal, geranyl acetate, nerol, geraniol, neral and traces of limonene and citral. Phytotherapeutic properties of Lemongrass oil are antimicrobial, antipyretic, antiseptic, astringent, bactericidal, deodorant, febrifuge, fungicidal, nervous system sedative and tonic. In addition, LGO also revitalizes the body and relieves the symptoms of jetlag, clears headaches and helps to combat nervous exhaustion and stress-related conditions. Traditional uses include its potential in severity of respiratory infections such as sore throats, laryngitis and fever and also conditions like colitis and gastro-enteritis. In this review, we present the scientific works published till the date. Formulation strategies, analytical works, bioactivities, toxicology and animal studies have been discussed in brief in this report.

KEYWORDS

Chromatography, *Acne vulgaris*, Mosquito-repellant, Anti-dandruff, Lemongrass and Hot infusion.

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INTRODUCTION

Cymbopogon is a genus of about 55 species of grasses, of which *Cymbopogon citratus* is native to warm temperate and tropical regions of Asia [1]. Lemongrass, citronella grass, fever grass, tanglad are the common names of *Cymbopogon citratus.* Lemongrass is native to India and tropical Asia. It is widely used as a herb in Asian cuisine. It has a subtle citrus flavor and can be dried and powdered, or used fresh [2]. Lemongrass oil is used as a pesticide and a preservative. Research shows that Lemongrass oil has anti-fungal properties. Lemongrass is said to act

as a fungicide when applied topically or to aid in digestion when taken as a tea. It can be purchased in certain markets or stores that cater to an Asian clientele, as it is used primarily as an ingredient in Thai cuisine. It is used in high fever, in clotting of blood in case of injuries. It also has anticancer properties and can be used as a muscle relaxant [1].

CHEMICAL COMPOSITION

The oil consists of certain natural chemical components including myrcene, citronellal, geranyl acetate, nerol, geraniol, neral and traces of limonene and citral (Table No.1). West Indian LGO contains a volatile oil (usually 0.2-0.4%), β-sitosterol, hexacosanol and triacontanol, cymbopogonol (orientin, isoorientin, isoscoparin, swertiajaponin, chlorogenic acid and caffeic acid and others) [3]. West Indian LGO contains citral (65–85%) as its major component. Cameroonian LGO contains geranial (33%) as the major component. Other compounds present include myrcene (12–20%), dipentene, methylheptenone, β-dihydropseudoionone, neral and others [3]. East Indian Lemongrass contains a volatile oil in about 0.5% yield from fresh grass. East Indian Lemongrass oil contains normally citral as its major component in a 70–85% concentration. Other components include geraniol and methyleugenol as well as many of the compounds present in the West Indian oil (dipentene, myrcene, methylheptenol, farnesol, n-decanal, guanic acid, and others) [4]. Geraniol-rich strains of East Indian Lemongrass have been reported to yield oils that contain citral only as a minor component (10–20%), with their major components being gerianol (35–50%) and methyl eugenol (20%). Another type is reported to contain no citral at all but has borneol (30%) as one of its major components [4]. East Indian Lemongrass oil usually contains a slightly higher content of citral than West Indian Lemongrass oil; it is also more soluble in 70% alcohol than the West Indian oil [5].

EXTRACTION PROCEDURE

Extracting oils from plant material can be done in several ways. Two popular methods, tincturing and hot infusion, result in a medicinal oil to be taken orally in hot tea or straight onto the tongue or as therapeutic, topical oil [3].

TINCTURE

To make tincture, the fresh Lemongrass stalks are broken and filled in the canning jar halfway (Figure No.1). Breaking the stalks allows the natural oils to be released from the plant and strengthen the tincture. The jar is half filled with alcohol and half with cold water. Vodka or brandy is the most common alcohols for making tinctures, but gin is used as well. Instead of alcohol, a half-and-half solution of white or apple cider vinegar and water is a suitable substitution. Then the lid is covered and the content is gently shaken with alcohol solution. Herbs are allowed to settle and monitored to see that all the plant material is covered by liquid. Even a small bit peeking out could mold during the tincturing process, ruining the tincture. Further they are placed in a cool, dark room for next three days. Thereafter, the solution is poured into a blender for greater absorption between plant material and the liquid, (because Lemongrass is such a woody plant). The blended liquid is again put back into the jar and stored for at least three weeks. After that, the content is squeezed and poured into a clean glass jar and stored until ready to use. Tinctures are commonly taken directly on the tongue but adding the medicine to a cup of warm tea or water may be a more palatable solution [3].

HOT OIL INFUSION

For making hot LGO infusion, extra virgin olive oil is poured into the top pan of a double boiler (Figure No.2). Half crushed stalk of Lemongrass is placed in the oil. Lemongrass should not be washed before putting in the pot. The content is heated in double boiler until the water on the bottom begins to steam. The Lemongrass is infused in the oil for at least an hour, but the longer the better. The infusion is cooled down and strained through a mesh strainer. This oil is used as a topical ointment or as soothing massage oil for variety of purposes [3].

ANALYTICAL METHODOLOGY

Some of the analytical works has been reported in literature elsewhere including UV, HPLC, HPTLC and Gas Chromatography method (Table No.2).

HIGH PERFORMANCE LIQUID CHROMATOGRAPHY

HPLC is the one the most significant methods for the analysis of oils in samples and matrix. In one of the study, the concentration of lemongrass oil was determined by HPLC at different doses of the oil (5-50 mg) and 5µl of the surfactant (25% Tween®-20 solution in sterile water) [6]. Aflatoxin residues were derivatized with trifluoro acetic acid (TFA) according to the method described by Dayananda (1990). The derivatised aflatoxin samples and AFB1 were analysed by reverse phase (C-18 column) HPLC system with a fluorescence detector. The solvent system was water: methanol: acetonitrile (60: 20: 20) with 360 nm and 445 nm excitation and emission filters. The flow rate was at 1 ml min^{-1}. Three replicates were injected (10 μ l) and the quantification of AFB_1 in samples was performed using the peak height [7].

In another study, a HPLC method for the quantitative determination of citral in *Cymbopogon citratus* volatile oil was developed and validated. The HPLC assay was performed using a Spherisorb® CN column (250 mm × 4.6 mm, 5 µm), a n-hexane:ethanol (85:15) mobile phase and an UV detector (set at 233 nm). The following parameters were evaluated: linearity, precision, accuracy, specificity, quantification and detection limits. The concentration of citral in C. citratus volatile oil obtained with this assay was 75%. The HPLC method developed in this study showed an excellent performance (linearity, precision, accuracy and specificity) and can be applied to assay citral in volatile oil [8].

HIGH PERFORMANCE THIN LAYER CHROMATOGRAPHY

According to various pharmacopeias, the quality control of essential oils and formulations containing vegetable oils or extracts from medicinal plants is very important and should involve analytical quantification of the constituents present in the sample, in particular the main component. Previously we have reported a HPTLC method for the analysis of citral isomers in Lemongrass oil. The HPTLC analysis was performed on a precoated Silica Gel Plates using toluene and ethyl acetate in 93:07 v/v as a mobile phase. HPTLC was performed to determine the percentage of main compound present in the oil [9]. Simultaneous assay of several components in a single formulation is possible by HPTLC [10].

GAS CHROMATOGRAPHY-MASS SPECTROSCOPY

GCMS is the method of choice for estimation of volatile oils and mentioned in various monographs. In one of the studies, essential oil was analyzed by using a Hewlett Packard 5890 II GC, equipped with a HP-5 MS capillary column (30 m × 0.25 mm i.d., 0.25 µm) and a HP 5972 mass selective detector. For GC-MS detection, an electron ionization system was used with ionization energy of 70 eV. Helium was the carrier gas, at a flow rate of 1 ml/min. Injector and MS transfer line temperatures were set at 220 and 290^{0}C, respectively. Column temperature was initially at 50^{0}C, then gradually increased to 150^{o}C at a 3^{o}C/min rate, held for 10 min and finally increased to 250^{o}C at 10^{o}C/min. Diluted samples (1/100 in acetone) of 1.0 µl were injected manually and splitless. The components were identified based on the comparison of their relative retention time and mass spectra with those of NBS75K library data of the GC-MS system, literature data and standards of the main components [10]. LGO was analyzed in a Shimadzu Gas Liquid Chromatograph coupled with a non - polar DB-5 capillary column [30 m x 0.250 mm and 0.25 micron internal diameter (ID)] using a flame ionization detector (FID). The percent of each compound was based on the peak area divided by the total area of component peaks. The temperature range was from 50-250^{0}C, with a temperature programme rate of 20^{0}C min -1, stating at two minutes and finishing at ten minutes. The detector and injection temperatures were both 250^{0}C. The retention indices (RI) of standard compounds that matched with RI of the unknowns that were detected by GC were identified. Citral was confirmed based on RI match with an authentic compound [12].

ETHANOBOTANICAL CONSIDERATION

Lemongrass belongs to the section of Andropogan called *Cymbopogan* (Family: Germineae) is a large perennial herb (Figure No.3). It has slightly branched partly aerial rhizome, reaching half inch in diameter and strongly ringed with the closely placed scars of the leaf-sheaths, the remains of which persist on the upper portion and giving off numerous tough fibres. Stems reaching 6 feet or more in height, erect and cylindrical. Leaves are very large and long, numerous erect lower ones sometimes reduced to their sheaths. It occurs in Himalayas from Kashmir to Assam ascending up to 10,800 feet. This beautiful grass is a native of Ceylon. Here it grows up to the level of 300 feet in well drained sandy soil. An annual rainfall of 203 to 254 cm and average temperature of 75 to 80 F are reported to be favorable in its growth. It is also cultivated in West Indies, Guatemala, Haiti and India and to a very limited extent in Pakistan.*C. Flexuosus Stapf.* is considered to have originated in Kerala [India]. The plant is very hardy and grows under a variety of conditions. The most ideal conditions are warm and humid climate with plenty of sunshine. Maximum age of Lemongrass is 18 to 24 months, while it is necessary to renew its plantation after every 6 to 8 years [13].

PHYTOTHERAPEUTIC POTENTIAL

Lemongrass oil is widely used to flavor Asian and Indian cuisine. Lemongrass is used as a traditional herbal folk remedy where lemongrass oil extractives are used by making an infusion or tea by pouring boiling water over the leaves of the lemongrass plant. Due to its common use in folk medicine, Lemongrass is bestowed with a plethora of medicinal properties which have not been scientifically proven [1]. Scientifically-based properties of Lemongrass oil include attributes of an antioxidant, insect repellent, analgesic, anti-inflammatory, antimicrobial and diuretic [14]. Believed to be helpful for cardiac symptoms, the utility of Lemongrass oil for cardiac disease sufferers is undetermined, but experiments have shown that it can reduce heart rates. Other reported, but largely undocumented uses for Lemongrass include medicinal uses against cancer and diabetes [2]. In dermal Pharmaceutics, Lemongrass oil is favored as a massage oil, and tonic for arthritis and circulatory system improvement. As with the other essential oils we have discussed, Lemongrass is used as a tonic for the nervous, respiratory, skin and digestive systems and may be used as a wound dressing. Lemongrass oil may also be used as a natural flea and tick remedy for household pets [15].

RELAXES MUSCLES

The most important health benefit of Lemongrass oil is its ability to calm and relax the muscles. It provides relief from headache, migraines and muscle strains [16].

DEPRESSION

Lemongrass oil is also useful in relieving symptoms related to depression, anxiety and panic attacks [16].

EDEMA

Its diuretic property is useful in curing edema and its carminative properties help relieve gas and acidity [17].

DIGESTION

Consumption of Lemongrass oil improves digestion and eases bowel movements in case of constipation [16].

FEVER

Lemongrass oil has antipyretic properties that helps reduce temperature during high fever and also eliminates fever completely [2].

INFECTIONS

The fungicidal, antimicrobial and antibacterial properties of Lemongrass oil make it beneficial in the prevention and cure of fungal as well as bacterial infections, both internal and external [2].

CLOTTING OF BLOOD

The astringent properties of Lemongrass oil help to speed up the process of blood clotting and prevent excessive loss of blood when applied to injuries that bleed profusely [16].

NERVE TONIC

Lemongrass grass oil can help cure many nervous system related disorders like Vertigo, Alzheimer's, Parkinson's disease, etc, by strengthening the nerves and the immune system as well [16].

ANTI-CANCER

Research indicates that Lemongrass or Lemongrass oil can cause programmed death in cancerous cells, without harming the useful cells [16].

SKIN CARE

Its antibacterial, astringent and antiseptic properties prevent and cure skin infection, prevent acne breakouts and soothe broken or inflamed skin. It is used as an ingredient in many skin care products like soaps, lotions, perfumes and deodorants [16].

CULINARY USES

Lemongrass oil is used in many continental recipes as an ingredient and garnish and also in various types of herbal teas and soups [16]. Lemongrass oil can also be used as an insecticide, disinfectant and for household cleaning and deodorizing purposes. It is also an effective mosquito repellent and a food preservative [16].

FORMULATION DEVELOPMENT

Table No.3 depicted a summary of formulations attempted on LGO. In one of the study, ointment and cream formulations of Lemongrass oil in different classes of base and the oil in liquid paraffin solution have been evaluated for mosquito repellency in a topical application [3]. Mosquito repellency was tested by determining the bite-deterrence of product samples applied on an experimental bird's skin against a 2-day starved culture of *Aedes aegypti* L. mosquitoes. Ointment preparations of the oil exhibited [greater than or equal to] 50% repellency lasting 2-3 h, which may be attributed to citral [3]. Another preparation of Lemongrass oil was anti-acne micro emulsion formulation of essential oil. We earlier studied the action of LGO formulation as microemulsion in case of *acne vulgaris*. In this study a natural compound was identified that has potential application in the treatment of *acne vulgaris* and a novel drug delivery system was developed for the same [18]. Pure essential oil and a topical formulation from Lemongrass oil (*Cymbopogon citratus*) Stapf have been investigated for repellent activity against *Acarus sacchari* and mosquitoes respectively. It was finally concluded that lemongrass oil is a promising natural repellent due to its safety advantage over chemical repellents [19]. In another study, the antifungal activity of Lemongrass oil against *Malassezia furfur*, opportunistic yeast associated with dandruff was evaluated by using a broth dilution assay and found effective in this regard [20]. LGO was also investigated for the pediculicidal activity with *Mentha piperita* oil. The pediculicidal potential of the formulation containing combined essential oils of *Cymbopogan citratus* and *Mentha piperita* oil was found to be excellent and the mean death time observed with this group was 64 minutes, which was comparable to positive control group (mean death time 53 minutes) [21].

RESULTS

Lemongrass from which lemongrass oil is made, encompasses a number of perennial, tall, tropical grass species found and cultivated in Central and South America and the West Indies. The plant is believed to have originated in Asia. Lemongrass oil is amber or dark yellow in color and has a scent that is both sweet and strong [22]. Lemongrass oil, although not related to citrus fruit essential oils, has many of the same properties and coincides with some similar uses of lemon and lime oils. Lemongrass oil is considered to have low toxicity in small doses, but its toxic properties are undetermined with regard to pregnant women and nursing children. Because of this lack of data, Lemongrass oil should not be used by pregnant or nursing mothers. Although not generally reactive, cases of sensitivity have been observed in some individuals and Lemongrass oil should not be inhaled, as this may cause lung damage. Like lemon and lime oil, Lemongrass oil contains citral which lends itself to the trademark citrus aroma of Lemongrass products [14]. The oral toxicity of Lemongrass oil in rats is more than 5 gm/kg i.e. Acute oral LD50 >5g/kg [23]. The LD 50 of citral which is a mixture of neral and geranial in rats is approximately 5gm/kg [23].

DISCUSSION

In the current scenario of increasing prevalence of obesity and diabetes, citral may prove as novel agent in its management. Currently, there is very little scientific evidence investigating the use of

Lemongrass in humans and more evidence is needed to make strong recommendations for its use as a sedative or for lowering high cholesterol. These uses have been tested in humans or animals. Safety and effectiveness have not always been proven. Some of these conditions are potentially serious, and should be evaluated by a qualified healthcare provider. Early research had not shown any effect of Lemongrass on serum cholesterol. However, more research is warranted in this area.

Table No.1. Phytoconstituents of Lemongrass Oil

S.No	Chemical Constituents	%Composition	Reference
1	Geranial	48.1%	[3]
2	Neral	34.6%	[3]
3	Myrcene	11.0%	[3]
4	Geraniol	1.9%	[3]
5	Linalool	0.7%	[3]
6	Limonene	2.46%	[4]
7	Citral	0.37%	[4]
8	Geranyl acetate	1.95%	[4]
9	Nerol	0.39%	[4]
10	Citronellol	0.44%	[4]

Table No.2: Analytical Methodology

S.No	Method	Conditions	Inference	Ref
1	HPLC	C-18 column, Solvent system: Water:methanol:acetonitrile(60:20:20) at 360 nm and 445 nm. Spherisorb®CNcolumn, mobile phase: n hexane**:**ethanol(85**:**15), an UV detector (set at 233 nm)	Aflatoxin residues in LGO were analyzed. Concentration of citral in LGO was analyzed.	[24,6] [7,8]
2	HPTLC	Mobile phase composition was Toluene: Ethyl acetate (93:07 v/v) and scanned at 595 nm	trans and cis-citral in Lemongrass oil were analyzed	[9,10]
3	GCMS	HP-5 MS capillary column, using Helium as carrier gas	LGO was analyzed for constituents	[11]
4	GC	DB-5 capillary column and FID detector	Citral was confirmed	[12]

Table No.3: Formulation Development

S.No	Formulation	Method	Purpose	Ref
1	Emulsion	Liquid paraffin solution	Mosquito-repellant	[3]
2	Microemulsion	Surfactants: Tween 80 and Tween 20 , Cosurfactants: Transcutol-P, Ethanol, and Distilled Water	*Acne vulgaris*	[17]
3	Cream	Liquid paraffin solution	Mosquito-repellant	[18]
4	Shampoo	Broth dilution assay	Anti-dandruff	[20]
5	Solution	Filter paper diffusion bioassay	Licicidal activity against human head lice	[21]

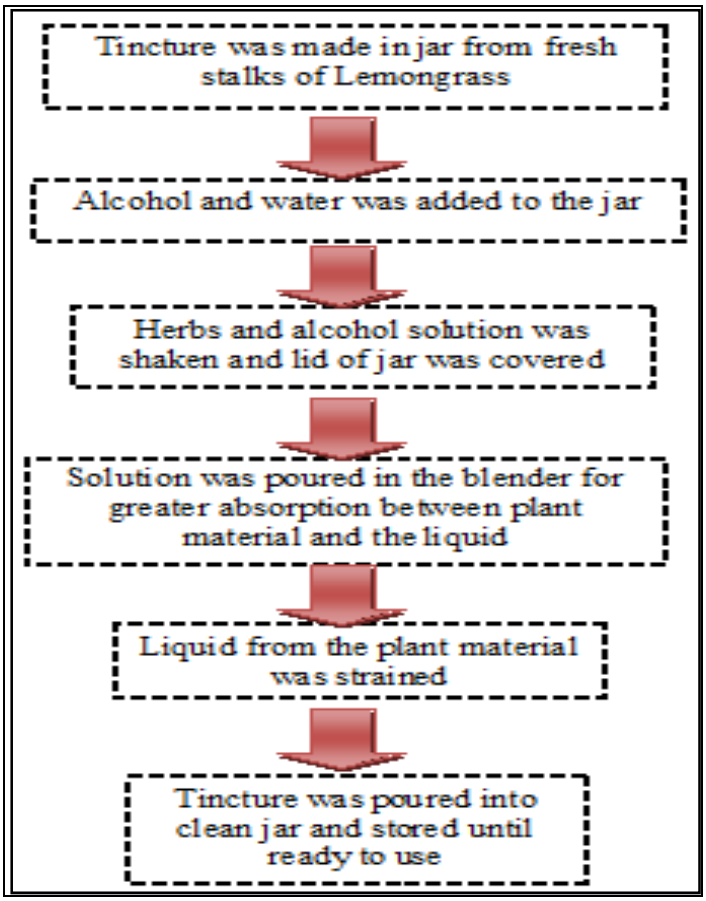

Figure No.1: Steps in Lemongrass Oil Extraction [3]

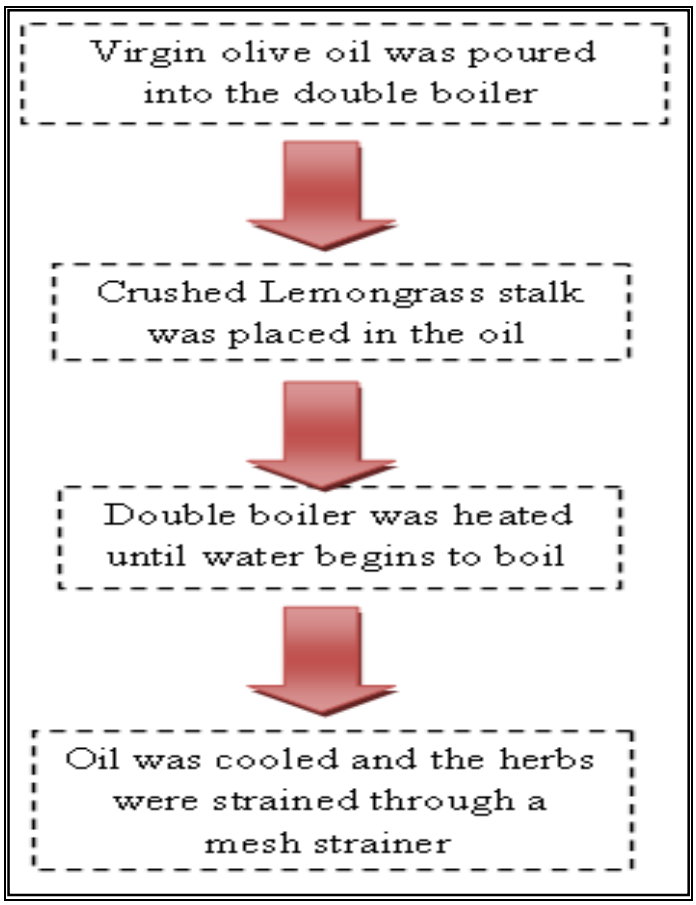

Figure No.2: Steps involved in Hot Oil Infusion [3]

Figure No.3: Exomorpic feature of Lemongrass [13]

CONCLUSION

Lemongrass oil is amber or dark yellow in color and has a scent that is both sweet and strong. Lemongrass oil is considered to have low toxicity in small doses. Lemongrass oil contains citral which lends itself to the trademark citrus aroma of Lemongrass products.

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