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Research Article

COMPARATIVE ANALYSIS OF ANTIOXIDANT AND FREE RADICALS SCAVENGING ACTIVITY OF LEMONGRASS OIL AND LAVENDER OIL

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ABSTRACT

Objective: To compare the free-radical scavenging activity of lemongrass oil and lavender oil.

Methods: Lemongrass oil and lavender oil were used to assess their antioxidant effect by *in vitro* methods such as phytochemical tests, nitric oxide radical scavenging activity, and estimation of total phenolic content (TPC).

Results: The oils were studied for different phytoconstituents and show the presence of carbohydrates, tannins, quinones, cardiac glycosides, terpenoids, phenols, and steroids in both the oils. These constituents can be used in the treatment of diarrhea, hemorrhage, cardiac failure, bacterial and fungal infections, dermatoses, burns and swollen joints, etc. The TPC of the oils were measured, and results show their use in anticancer and antioxidant effect. The nitric oxide assay of two oils shows their scavenging activity and their use in controlling blood pressure, platelet functions, etc.

Conclusion: Both the oils show great antioxidant effect. They can be used as a medicinal supplement due to its natural availability and its use in cancer treatment.

Keywords: *Cymbopogon citratus*, *Lavandula angustifolia*, Phytochemicals, Free radical scavengers, Antioxidants.

INTRODUCTION

Cymbopogon citratus (lemongrass) is a perennial tropical grass and it is resistant to different temperatures and can grow in warm, semi-warm, and temperate climates. It is from 60 to 120 cm high; its leaves are green, long and slats and have pleasant aroma and taste. This grass is native to India [1]. The leaf essential oil is used in the food, perfumery, soap, cosmetic, pharmaceutic, and insecticide industries. The main constituents of this essential oil are citral (aldehyde geraniol+neral) and terpenes (myrcene-monoterpene) and geraniol-terpenic alcohol). The comprehensive account of the chemical constituents and the biological activities are presented in this research is to allow an evaluation of the potential use of this plant either in pharmaceutic or as an agricultural resource [2]. Various pharmacological activities of *C. citratus* have been reported such as antiamoebic, antibacterial, antidiarrheal, antifungal, antimalarial, antiinflammatory, and antianxiety. The tea made from its leaves is popularly used as antispasmodic, analgesic, antiinflammatory, antipyretic, diuretic, and sedative. A few ethnobotanical reports on the treatment of fever and headache were investigated [3]. The genus Lavandula is native to the lands surrounding the Mediterranean Sea and Southern Europe through Northern and Eastern Africa and Middle Eastern countries to Southwest Asia and Southeast India. It includes more than 30 species, dozens of subspecies, and hundreds of hybrids and selected cultivars. The different varieties of this plant range in height from 9 inches to 3 feet, although some may grow taller with age. The main constituents of lavender are linalool, linalyl acetate, 1,8-cineoleB-ocimene, terpinen-4-ol, and camphor [4]. Lavender essential oil extracted from lavender (*Lavandula angustifolia*) is popular as a complementary medicine in its own right and as an additive to many over the counter complementary medicine and cosmetic products. It is used as a therapeutic agent as well as an antibacterial agent for centuries. It is believed to have sedative, carminative, antidepressive, and antiinflammatory properties [5]. It is found to be beneficial in the fields of dermatology, gastritis, respiratory complaints, wound healing, and genital infections. Of all the essential oils used commercially, lavender oil appears to be one of the most popular [6]. In recent years, several animal and human investigations have indeed evaluated traditional medical remedies of lavender using modern scientific methods. These studies raised the possibility of revival of lavender therapeutic efficacy in neurological disorders on the basis of evidence-based medicine [7,8]. Free radicals are produced in normal or pathological cell metabolism. Oxidation is essential to many living organisms for the production of oxygen-derived free radicals is involved in triggering many diseases such as cancer, rheumatoid arthritis, cirrhosis, and arteriosclerosis as well as in degenerative processes associated with aging. Exogenous chemical and endogenous metabolic processes in the body or in the digestive system might produce highly reactive free radicals, especially oxygen derived radicals which are capable of oxidizing biomolecules resulting in cell death and tissue damage [9]. Almost all the organisms are well protected against free-radical damage by oxidative enzymes such as superoxide dismutase and catalase, or by chemicals such as a-tocopherol, ascorbic acid, carotenoids, polyphenols, and glutathione [10]. When the process of an antioxidant protection becomes unbalanced, deterioration of physiological functions may occur, resulting in accelerated aging dieases. Antioxidant food supplements may be used to help the human body to reduce oxidative damage. Natural antioxidants are being extensively studied for their capacity to protect organisms and cells from damage brought on by oxidative stress [11]. The use of essential oils as functional ingredients in foods, drinks, toiletries and cosmetics is becoming popular [12]. The aim of this research is to conduct a comparative evaluation of antioxidant properties of lemongrass oil and lavender oil.

METHODS

The chemicals and reagents used in this research are obtained from Himedia.

Phytochemical tests

Phytochemical analysis was done as per Harborne and Kokate.

Test for carbohydrates

To 2 ml of plant extract, 1 ml of Molisch's reagent and few drops of concentrated sulfuric acid were added. The presence of purple or reddish color indicates the presence of carbohydrates [13].

Test for tannins

To 1 ml of plant extract, 2 ml of 5% ferric chloride was added. Formation of dark blue or greenish black indicates the presence of tannins [14].

Test for saponins

To 2 ml of plant extract, 2 ml of distilled water was added and shaken in a graduated cylinder for 15 minutes lengthwise. Formation of 1 cm layer of foam indicates the presence of saponins [15,16].

Test for flavonoids

To 2 ml of plant extract, 1 ml of 2 N sodium hydroxide was added. The presence of yellow indicates the presence of flavonoids [17].

Test for alkaloids

To 2 ml of plant extract, 2 ml of concentrated hydrochloric acid was added. Then, few drops of Mayer's reagent were added. Presence of green or white precipitate indicates the presence of alkaloids [18].

Test for quinones

To 1 ml of extract, 1 ml of concentrated sulfuric acid was added. Formation of red color indicates the presence of quinones [19].

Test for glycosides

To 2 ml of plant extract, 3 ml of chloroform and 10% ammonia solution was added. Formation of pink indicates the presence of glycosides [20].

Test for cardiac glycosides

To 0.5 ml of extract, 2 ml of glacial acetic acid and few drops of 5% ferric chloride were added. This was under layered with 1 ml of concentrated sulfuric acid. Formation of brown ring at the interface indicates the presence of cardiac glycosides [21].

Test for terpenoids

To 0.5 ml of extract, 2 ml of chloroform was added and concentrated sulfuric acid was added carefully. Formation of red-brown at the interface indicates the presence of terpenoids [17].

Test for phenols

To 1 ml of the extract, a few drops of Phenol Ciocalteau reagent was added followed by few drops of 15% sodium carbonate solution. Formation of blue or green indicates the presence of phenols [19].

Test for coumarins

To 1 ml of extract, 1 ml of 10% NaOH was added. Formation of yellow indicates the presence of coumarins [19].

Steroids and phytosteroids

To 1 ml of plant extract, equal volume of chloroform is added and subjected with few drops of concentrated sulfuric acid appearance of brown ring indicates the presence of steroids and appearance of bluish-brown ring indicates the presence of phytosteroids [21].

Phlobatannins

To 1 ml of plant extract, few drops of 2% HCL was added the appearance of the red precipitate indicates the presence of phlobatannins [17].

Anthraquinones

To 1 ml of plant extract, few drops of 10% ammonia solution was added, the appearance pink precipitate indicates the presence of anthraquinones [17].

Estimation of total phenolic contents (TPC)

TPC of extracts was assessed according to the Folin–Ciocalteu method [22] with some modifications. Briefly, different concentrations of oils (200, 400 and 600 µg), made to 2 ml with distilled water and 1 ml of Folin–Ciocalteu's reagent was seeded in a tube, and then 1 ml of 100 g/l Na_2CO_3 was added. The reaction mixture was incubated at 25°C for 2 hrs and the absorbance of the mixture was read at 765 nm. A calibration curve with six data points for catechol was obtained.

Antioxidant assay

Nitric oxide radical inhibition assay

Sodium nitroprusside in an aqueous solution at physiological pH spontaneously generates nitric oxide; it interacts with oxygen to produce nitrite ions, which can be estimated using GriessIllosvoy reaction [23]. In the present investigation, GriessIllosvoy reagent was modified using naphthylethylenediaminedihydrochloride (0.1% w/v) instead of 1-naphthylamine (5%). The reaction mixture (3 ml) containing sodium nitroprusside (10 mM, 2 ml), phosphate buffer saline (0.5 ml) and different concentration of oils (200–600 µg) or standard solution (0.5 ml) were incubated at 25°C for 150 min. After incubation, 0.5 ml of the reaction mixture containing nitrite was pipetted and mixed with 1 ml of sulphanilic acid reagent (0.33% in 20% glacial acetic acid) and allowed to stand for 5 min for completing diazotization. Then 1 ml of naphthylethylenediaminedihydrochloride (1%) was added, mixed and allowed to stand for 30 min. A pink coloured chromophore was formed in diffused light. The absorbance of these solutions was measured at 540 nm against the corresponding blank [23]. Ascorbic acid was used as positive control. The scavenging activity was calculated using the formula.

% of Inhibition = (A of control–A of test)/A of control*100

RESULTS AND DISCUSSION

In this research, antioxidant effect of two medicinal plants such as lemongrass oil and lavender oil is measured using *in vitro* methods such as photochemical tests, nitric oxide radical inhibition assay, and estimation of TPC.

Phytochemical tests

The extracts were studied for phytoconstituents such as carbohydrates, tannins, saponins, flavonoids, alkaloids, quinones, glycosides, cardiac glycosides, terpenoids, phenols, coumarins, steroids and phytosteroids, phlobatannins, and anthraquinones by different phytochemical tests. The results were revealed the presence of carbohydrates, tannins, quinones, cardiac glycosides, terpenoids, phenols and steroids in both the oils. Saponins present weakly in these oils (Table 1).

The presence of these constituents' acts as antioxidants and helps in scavenging free radicals. They can be used as oxidative stress ailments. The tannins have been used for the treatment of diarrhea,

Table 1: Presence and absence of phytoconstituents in lemongrass oil and lavender oil

Serial number	Phytochemical tests	Lemongrass oil	Lavender oil
1	Carbohydrates test	+	+
2	Tannins test	+	+
3	Saponins test	Weakly +	Weakly +
4	Flavonoids test	–	–
5	Alkaloid test	–	–
6	Quinones test	+	+
7	Glycosides test	–	–
8	Cardiac glycosides test	+	+
9	Terpenoids test	+	+
10	Phenols test	+	+
11	Coumarins test	–	–
12	Steroids and phytosteroids	Steroids	Steroids
13	Phlobatannins test	–	–
14	Anthraquinones test	–	–

Table 2: Phenolic content of the lemongrass oil and lavender oil

Concentration (µg)	Lemongrass oil	Lavender oil	Catechol	Phenol content of the lemongrass oil	Phenol content of lavender oil
200	0.0432	0.0394	1.844	4.685466377	4.273318872
400	0.0741	0.0791	3.565	8.314165498	8.875175316
600	0.0959	0.0996	5.552	10.36383285	10.76368876

Table 3: Nitric oxide scavenging activity of lemongrass oil and lavender oil

Concentration (µg)	Control	Lemongrass oil	Lavender oil	Ascorbic acid	Percentage of lemongrass oil	Percentage of lavender oil	Percentage of ascorbic acid
200	0.784	0.502	0.528	0.405	35.96938776	32.65306122	48.34183673
400	0.784	0.385	0.402	0.246	50.89285714	48.7244898	68.62244898
600	0.784	0.254	0.286	0.118	67.60204082	63.52040816	84.94897959

hemorrhage and detoxification [24]. Saponins help in control of blood cholesterol levels, bone health, cancer, and building up the immune system. Quinones show pharmacological activities such as antitumor, antimicrobial, antiparasitic, and anti-cardiovascular diseases. They can also be used as a purgative. Cardiac glycosides have been adapted for the treatment of congestive heart failure and cardiac arrhythmia. It primarily involves in the treatment of cardiac failure. Terpenoids have been used for viral, bacterial and fungal infections, dermatoses, burns, and swollen joints. Phenols are antiseptic and antipruritic in nature. Hence, all these constituents present in these oils has a great medicinal use which can utilized for the pharmacological application.

TPC

A calibration curve with six data points for catechol was obtained. The results were compared to a catechol calibration curve, and the TPC of extracts was expressed as mg of catechol equivalents per gram of extract. The results were tabulated (Table 3).

The TPC in both the oils is almost equal without showing any significant differences. Phenolic compounds in plants help in antioxidant effect, and it is mainly used for the treatment of cancer. Hence, these oils can be used as a preventative measure for cancer.

Antioxidant assay

Nitric oxide radical inhibition assay

Nitric oxide acts as a regulatory molecule and is the free radical generated from sodium nitroprusside in aqueous solution. Nitrite, one of the oxides of oxygen, was inhibited by plant oils in competition with other oxides in the reaction medium [24]. Both oils show the nitric oxide scavenging effect. The result of nitric oxide scavenging activity of two oils is illustrated in Table 3.

Clearly both these oils have the antioxidant activity. The scavenging activity of lemongrass oil is higher than lavender oil. However, the standard ascorbic acid has been highest scavenging activity among three. Nitric oxide has effects in controlling blood pressure, neural signal transduction, platelet function, antimicrobial, and anti-tumor activity.

CONCLUSION

From this research, it is proven that both lemongrass oil and lavender oil has an antioxidant effect. Nowadays, individuals tend to follow an unhealthy diet pattern. They prefer chemicals and drugs over natural remedies, which are harmful to the body. Naturally available plants extracts and their oils have many useful effects. The lemongrass oil and lavender oil are greatly useful for stress, anxiety, headache, diarrhea, etc., which are the common problems faced by all. They can also be useful as an anti-tumor, antimicrobial, antiparasitic, and anti-cardiovascular diseases. Its benefit as an antioxidant has the higher effects on free radicals scavenging activities, which can be used in preventing heart-related diseases and accelerated ageing. It is also useful in the treatment of cancer which is a growing threat faced by many people. All these medicinal effects can be utilized in the pharmacological application. Since these are oils, they can be taken externally in the case of burns, swollen joints and dermatoses as it contains terpenoids. As a whole, both these oil has medicinal benefits which can be utilized for the future ailments.

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