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PHYTOCHEMICAL SCREENING QUALITATIVE AND QUANTITATIVE ANALYSIS OF LEMON GRASS

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Abstract: Medicinal plants are the source of treatment for many diseases and ailments throughout the developing world. Studies on them could lead to the finding of novel drugs for effective treatment of various diseases. . We have chosen one of the most commonly used plants of *Cymbopogon citrates* leaves for the present study which was screened for phytochemical and antibacterial properties. Studies on them could lead to the finding of novel drugs for effective treatment of various diseases. We have chosen one of the most commonly used plants of *Cymbopogon citrates* leaves for the present study which was screened for Phytochemical analysis revealed the presence of several bioactive compounds such as flavonoids, phenols, tannins, alkaloids etc. Plants synthesize a vast variety of chemical compounds classified as primary and secondary metabolites. Primary metabolites are involved directly in growth and development whereas secondary metabolites have several medicinal importance. There are wide range of secondary metabolites such as alkaloids, flavonoids, saponins, tannins, terpenoids, cardiac glycosides etc. Each of these have specific functions and health benefits. Hence they are used as raw materials for pharmaceutical and cosmetic industries. The study revealed the medicinal potential of lemongrass.

Key words: Phytochemical; **Qualitative and Quantitative.**

I. INTRODUCTION

Lemongrass is a plant in the grass family. There are over 100 lemongrass species, including *Cymbopogon citratus*, which is often used in foods and medicine. Lemongrass leaf and essential oil contain chemicals that might help prevent some bacteria and yeast from growing. *Cymbopogon citratus,* commonly known as Lemongrass belongs to the poaceae family and genus Cymbopogon is a tall, monocotyledonous aromatic perennial plant with slender sharpedge green leaves with pointed apex. The origin of the plant is tropical Asia. Taxonomic details of the lemongrass: **(Gupta *et al.,* 2019).**

Kingdom : Plantae
Division : Magnoliophyta
Class : Liliopsida
Order : Poales
Family : Poaceae
Genus : Cymbopogon
Species : citrates

Lemongrass (*Cymbopogon citratus*) an aromatic herb, known in the North and West tropical Africa, in Arabian Peninsula and in Egypt, it was a native of (Southwest Asia) South Indiabut present, in many parts of the world growing in dense clumps. In the folk medicine of Brazil and Mexico (Uraku, 2015). *Cymbopogon citratus* staff were popularly known as citronella grass or lemongrass, this species belongs to the Gramineae family, which comprises approximately 500 genus and 8,000 herb species, the leaf-blade is linear, tapered at both ends and can grow to a length of 50 cm and width of 1.5 cm (**Manvitha & Bidya, 2014).**

MEDICINAL PLANTS: PROPERTIES AND APPLICATIONS

Lemon grass good for an effective antibacterial and antifungal agent that contains anti-inflammatory and antioxidant properties. Lemongrass contains quercetin, a flavonoid known for having antioxidant and anti-inflammatory benefits. Quercetin reduces inflammation, which inhibits cancer cell growth and prevents heart disease.Relieving anxiety. Many people find sipping hot tea to be relaxing, but lemongrass tea may offer further anxiety-reducing properties.,Lowering cholesterol. ,Preventing infection.Boosting oral health. Relieving pain,Boosting red blood cell levels.Relieving bloating. **(Hassan *et al., 2023).***

Lemongrass leaf and essential oil contain chemicals that might help prevent some bacteria and yeast from growing. Lemongrass also contains chemicals that might relieve pain and swelling. People use lemongrass for stomach pain, dandruff, high cholesterol, gingivitis, thrush, and many other conditions, but there is no good scientific evidence to support these uses. The plant is used as a fragrance and flavoring agent and in folk medicine as an antispasmodic, hypotensive, anticonvulsant, analgesic, antiemetic, antitussive, antirheumatic, antiseptic and treatment for nervous and gastrointestinal disorders and fevers. The plant is also used as an antibacterial, antidiarrheal and antioxidant. (***Shah Ge et al.,* 2011).**

SECONDARY METABOLITES

Plants synthesize a vast variety of chemical compounds classified as primary and secondary metabolites. Primary metabolites are involved directly in growth and development whereas secondary metabolites have several medicinal importance. There are wide range of secondary metabolites such as alkaloids, flavonoids, saponins, tannins, terpenoids, cardiac glycosides etc. Each of these have specific functions and health benefits. Hence they are used as raw materials for pharmaceutical and cosmetic industries . India has tremendous variety of plant sources and is origin of traditional system of medicines

Lemongrass (*Cymbopogon citrates*) plant leaves contained sufficient amounts of phytochemicals (alkaloids, glucosides, phenols, saponins, flavonoids, tannins, terpenoids and resins), steroids are absent. Chemicals contents of Lemongrass leaves are (Moisture, ash, fat, fiber, protein and Carbohydrate). (**Khalifah *et al.*, 2021).**

Phytochemicals are biologically active naturally occurring secondary metabolites found in vegetables, fruits, medicinal plants, aromatic plants, leaves, flowers and roots. They are responsible for defending the plants against disease environmental stress, UV exposure etc. along with imparting colour, fragrance and flavour to the plant. The medicinal value of plant lies in the bioactive phytochemical constituents of the plant.

II. MATERIALS AND METHODS

The plant material Fresh The fresh leaves of **Lemongrass (*Cymbopogon citrates*)** free from disease were collected from local nursery , Mehsana -384001,Gujarat.

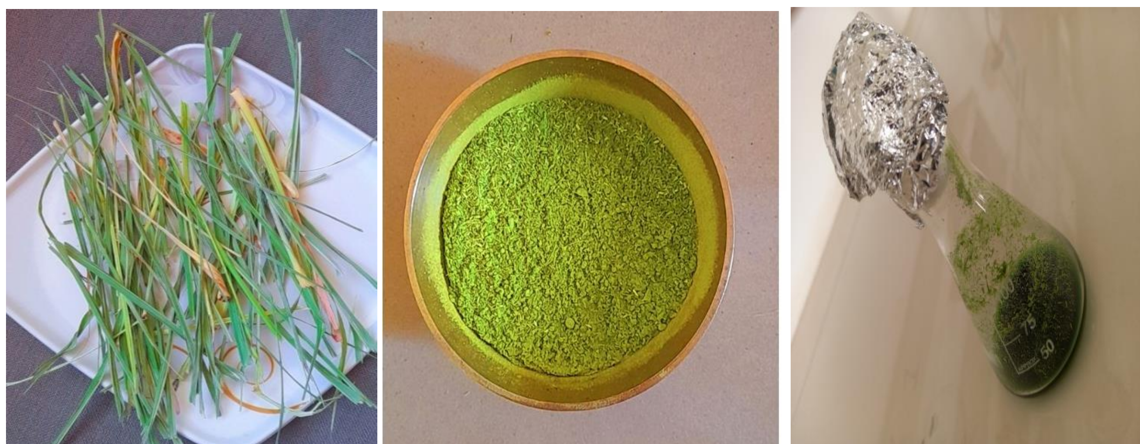

Figure 1: Plant sample preparation.

SAMPLE PREPARATION:

The collected sample was washed with running tap water to remove the dust particles and other contaminants present on the sample. The clean sample was once again washed with distilled water to avoid any cross contamination of the sample and the sample was air dried under the sun for 8-10 days at 27-30 degree Celsius and then subjected to electrical grinder to obtain fine powder. The powder was then subjected to sieving to obtain fine powder so that the extraction can be carried out at larger surface area . **(Nyamath *et al.,* 2018).**

METHODS:

The powdered plant samples (50g/250ml) were extracted succevely with Methanol and Distilled water and shaking 100 rpm for 15 minutes. The solution was allowed to settle down at room temperature for 24 hours. For each solvent of the respective extracts were reduced under room temperature and stored at 4˚C For further use. The dried plant extracts were then redissolved in dimethylsulfoxide (DMSO) and to get the solution of 10mg/ml for each extract which was subjected to analysis of in vitro antioxidant activities**.**

Figure 2: DMSO method for a plant extraction.

III. QUALITATIVE ANALYSIS OF PHYTOCHEMICALS

The quantitative analysis is very essential to identify the phytochemical constituent present in plants. The medicinal value of plants is due to the presence of particular bioactive constituents which can be easily accessed by standard methods.

Table.1: List of Qualitative test.

Sr. No	Qualitative test	Chemicals used	Interpretation	Reference
1	Saponin	1ml of extract add 1ml of D/W & shake vigorously	formation of the foam indicates the presence of saponins	(Tiwari *et al.*, 2016)
2	Alkaloids	Five milliliters of dilute ammonia solution was 3added to a portion of the aqueous filtrate of the extract followed by the addition of concentrated H_2SO_4.	Development of yellow color indicated the presence of alkaloids. The yellow coloration disappeared on standing.	(Tiwari *et al.*, 2016)
3	Steroids	1 ml of the extract was dissolved in 10 ml of the chloroform and equal volume of sulphuric acid was added by the sides of the test tube. If the upper layer turns red and sulphuric acid layer showed yellow with green fluorescence	If the upper layer turns red and sulphuric acid layer showed yellow with green fluorescence This indicates the presence of steroids.	(Tiwari *et al.*, 2016)
4	Phenolic compound	Ferric chloride test were carried out where the extract were diluted to 5 ml with distilled water. To this, a few drops of neutral 5 % ferric chloride solution was added.	A dark green or a blue black coloration indicated the presence of phenolic compounds.	(Tiwari *et al.*, 2016)
5	Flavanoid	Five milliliters of dilute ammonia solution was added to a portion of the aqueous filtrate of the extract followed by addition of concentrated sulfuric acid (H_2SO_4).	Appearance of yellow color indicated the presence of Flavonoid. The yellow coloration disappeared on standing.	(Tiwari *et al.*, 2016)
6	Tannin	The Folin-Denis colorimetric method was used to determine the presence of Tannin. About 0.5g of the dried powdered sample was taken in 20ml test tube and boiled for a minute then filtered through Whatman No. 42 filter paper. A few drops of ferric chloride were added.	A brownish green or a blue-black coloration indicated the presence of tannins.	(Ezeonu & Ejikeme 2016)
7	Glycoside	Extract were first hydrolyzed with dil. HCl and then subjected for glycoside analysis. Extracts were treated with Ferric chloride solution and immersed in boiling water for about 5 minutes. The mixture was allowed to cool and extracted with equal volume of benzene. The benzene layer was separated and treated with ammonia solution.	Formation of rose pink color in the ammonia layer indicates the presence of glycosides.	(Tiwari *et al.*, 2016)
8	Coumarins	0.5 ml of extracts was taken in test tubes. The mouth of the tube was covered with filter paper treated with 1 N NaOH solution. After that the test tubes were placed in boiling water and then filter paper was removed. Test tubes were examined under UV light.	Development of yellow fluorescence indicated the presence of coumarins	(Kumar *et al.*, 2013)

9	Quinines	Each 1ml of filtered sample was added with 1ml of sodium hydroxide.	The formation of blue, green, or red colors shows the presence of quinines.	(Tiwari *et al.*, 2016)

IV. QUANTITATIVE ANALYSIS OF PHYTOCHEMICALS

The quantitative analysis of phytochemical present in extracts was determined by standard procedures. All the extracts showed different amount of phytochemical. On quantification, among the six components, free amino acid content was highest in all the extracts. During the study Visible Spectrophotometer (Zeal-Tech 09171) was used to analyze the absorbance values of different phytochemicals.

Determination Total phenolic content

The method involves the reduction of folin-cioculteau reagent by phenolic compounds with a concomitant formation of the blue complex. In this study 0.5 gm dry gallic acid, 10ml ethanol and dilute to 100ml with distilled water can be stored at two weeks phenol concentration mg/L Gallic acid 50, 100, 150, 250, 500 each flask in gallic acid stock solution 1,2,3,5,10 ml and adjust the 100ml distilled water this stock solution in five different test tube in 200µl add and 1ml of Fe reagent add. One tube is sample in 200µl and 1ml Fe reagent added. After 40°C for 3min incubation in dark condition. After O.D at 765 nm in U.V spectrophotometer **(Blainski *et al*., 2013).**

Total alkanoid content

The total alkaloid content present in plants was determined by using UV spectrophotometer. This method is base on the reaction between alkaloid and bromocresol green. The part of the plant extract was dissolved in 2 N HCl and then filtered. 1 ml of this solution was transferred to separatory funnel and washed with 10 ml chloroform. The pH of the phosphate buffer solution was adjusted with 0.1N NaOH. 1 ml of this solution was transferred to a separating funnel and then 5 ml of bromocresol solution along with 5 ml of phosphate buffer were added. The mixture was shaken and the complex formed was fractioned with chloroform by vigorous stirring. The fractions were collected in a 10 ml volumetric flask and diluted to volume with chloroform. The absorbance of the complex in chloroform was measured at 470 nm. All the experiments were performed thrice (**Hikino *et al*., 1984**).

Total protein

1 gram of powder was taken in a test tube then 2 ml of phosphate buffer was added and mixed it properly. This sample was kept overnight for complete extraction of protein. This sample was centrifuged at 6000 rpm for 15 minutes. The supernatant is used for protein analysis and the pellet is discarded. For the quantitative analysis of protein, 5 ml of alkaline copper sulfate reagent was added to 1 ml of supernatant and mixed thoroughly. The solution was allowed to stand for 10 minutes and then 0.5ml of Folin's reagent was added. The tubes were incubated at room temperature for 30 minutes to develop color. The absorbance was recorded at 660 nm, against a blank. The blank is prepared by taking 1 ml of 0.5 M NaOH in place of sample in cuvette. Bovine serum albumin is used to plot standard curve (**Shah *et al.*, 2013**).

Total carbohydrate

The total soluble carbohydrate present in peel extract was determined according to the method described by **Hegde and Hofreiter**. 1 ml of sample was mixed with 4 ml of anthrone reagent. The tubes were incubated in boiling water bath for 8 minutes after which the absorbance was recorded at 630 nm against a reagent blank.

Glucose was taken to draw standard curve. The analysis was carried out in triplicate lates kept in refrigerator at 4-8 ^{0} temperature for 20 min. and then plates were incubated at 37°C for 18–24 hours. The zones of inhibition were measured.

V. RESULT AND DISCUSSION.

Table.2: Results for qualitative analysis of phytochemicals for *Cymbopogon citrates.*

Sr No	Test	d/w	Methanol
1	Saponin	-	-
2	Tannins	+	+ + +
3	Alkaloid test	-	+ + +
4	Steroid	-	+ +
5	Flavonoids test Sodium hydroxide test	-	+
6	Flavonoids test Lead acetate test	-	+
7	Glycoside	+	+
8	Quinones	-	-
9	Coumarin	-	-
10	Phenol	+	+

Figure.3: Results for qualitative analysis of phytochemicals for *Cymbopogon citrates* Methanol extract.

Figure.4: Results for qualitative analysis of phytochemicals for *Cymbopogon citrates* Distilled water extract.

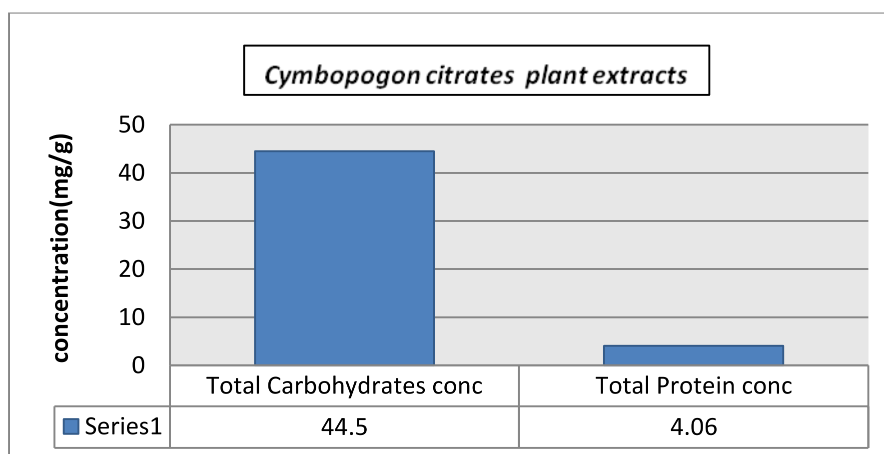

Figure.5: Results for Quantative analysis of phytochemicals for *Cymbopogon Citrates.*

DISCUSSION:

The phytochemical analysis was carried out using different solvents such as alkaloids, flavonoids, saponins, tannins, terpenoids, cardiac glycosides etc. The obtained results show the presence of Tannin, Saponin Tannins Alkaloid test Steroid Flavonoids test Glycoside in methanolic extract to comapair distilled water extract**.** In the extracts, Generally, Alkaloids, Steroid and tannins are responsible for the antioxidant property of the plants. Phenol, Alkaloid has been reported as bactericidal, antispasmodic and analgesic agent (**Gracelin*et et al.,* 2013; Velumani 2016**). We obtained the best results for the methanolic extracts followed by , distilled water. the plant extracts were subjected to quantification of phytochemical compounds. while we got negative results for Saponin , Coumarin and Quinones in distilled water and methanol extract.

VI. CONCLUSION

In the present study, medicinal plant **Lemongrass** (***Cymbopogon citrates***) was used to study their phytochemical analysis. There is lots of evidence suggesting that medicinal plants are very effective in the treatment of infectious diseases. The plants hold great promise as a source of novel antimicrobial agents. They are readily available, cheap and also, almost; do not have any side effects. However, many studies still need to be conducted to ensure the mechanism of action and also the safety of antimicrobial phytochemicals.

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