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Formulation and Development of Antibacterial Incense Sticks using Lemongrass oil

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ABSTRACT-
Lemongrass oil, known for its aromatic and therapeutic properties, is extracted from Cymbopogoncitratus using various techniques.Extraction of oil was performed using maceration and Soxhlet methods, with maceration yielding higher bioactivity. Qualitative phytochemical analysis of the extract revealed the presence of flavonoids, saponins, cardiac glycosides, terpenoids, quinones, coumarins, glycosides, and volatile oils. Anti-inflammatory activity of the essential oil was assessed through proteinase inhibitory assay showed that the maceration extract achieved higher inhibition, whereas Soxhlet extracts demonstrated lower inhibition. In vitro antibacterial assays using the broth dilution method indicated significant bacterial growth inhibition by maceration extracts against Bacillus subtilis. The obtained essential oil was then utilized in the development of incense sticks, integrating it with a base mixture of wood powder, charcoal, and a binding agent. The resulting incense sticks were evaluated for fragrance retention, burning time, and consumer acceptability. Lemongrass-based incense sticks were successfully formulated, exhibiting a 99% inhibition rate in fumigation studies, confirming strong antimicrobial activity. This research highlights the potential of lemongrass oil as a natural ingredient in incense production, offering a sustainable and eco-friendly alternative to synthetic fragrances while leveraging its antimicrobial and mood-enhancing properties.

Keywords-Cymbopogoncitratus, Phytochemical analysis, Anti- inflammatory activity, Incense sticks, Antimicrobial assay

I. INTRODUCTION-

Lemongrass (Cymbopogoncitratus)(Fig. 1) is a tropical grass known for its strong lemon scent due to high citral content. It has calming and insect-repelling properties and is commonly used in herbal tea. Lemongrass oil, a major product of India, is valued for its health benefits, including relieving muscle pain, inflammation, and fever. It also aids digestion, reduces stress, and helps treat diabetes and nervous disorders by boosting the parasympathetic nervous system (kulkarni, 2009; Boukhatem, 2014).

Fig. 1: Plant of Lemongrass

Lemongrass (Cymbopogon species), also known as citronella grass, is a tropical plant from the Poaceae family, found in warm regions of Asia, America, and Africa, particularly in India, Sri Lanka, and Southeast Asia. It is easy to grow, profitable for farmers, and in high demand for local use and export in cooking, medicine, cosmetics, and cleaning products.(Onoriode, 2023).Lemongrass is classified as follows:

- **Kingdom**: Plantae
- **Clade**: Tracheophytes
- **Clade**: Angiosperms
- **Clade**: Monocots
- **Clade**: Commelinids
- **Order**: Poales
- **Family**: Poaceae
- **Subfamily**: Panicoideae
- **Genus**: Cymbopogon
- **Species**: Cymbopogoncitratus(Nambiar, 2012)

Lemongrass (Cymbopogon species) contains a mix of volatile essential oils and non-volatile phytochemicals, including aldehydes, alcohols, terpenes, phenolic compounds, and

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nitrogenous compounds, each contributing to its medicinal and industrial uses. (Onoriode, 2023)

Lemongrass species include Cymbopogoncitratus, Cymbopogonnardus, and others. It grows in clumps up to 6 feet tall with long green leaves and a sturdy stem. Its essential oil, rich in citral (over 70%), is used in perfumes, soaps, insect repellents, Ayurvedic medicines, and flavoring agents. India is the leading producer, supplying nearly 80% of the world's lemongrass oil. It can be harvested up to five times a year, making it a valuable crop with both local and export markets (Onoriode, 2023)

In Asia, South America, and Africa, people have long used lemongrass for: Tea, Deodorants, Repellents.

Lemongrass is popular in cooking, especially in Asian cuisines. Its uses include: Flavoring teas, soups, and curries, and preparing a paste for cooking. Making lemongrass iced tea or a simple lemongrass syrup for drinks and desserts. Enhancing the taste of meats, seafood, sauces, wines, and baked goods (Onoriode, 2023).

Lemongrass has many health benefits. It can be used fresh, dried, or as an oil and helps with: Nausea, stomach aches. constipation and digestive issues. Reducing inflammation and fevers.Fighting bacteria and pests.

Phytochemical properties are the physical and chemical characteristics of the substance or species which define its behaviour under various conditions. It include color, odour, specific gravity, refractive index, optical reaction, solubility, acidic value, boiling point, moisture content, etc.

II. MATERIALS AND METHODS-

2.1 Collection of Sample

Lemongrass was collected from the market(vegetable market, pune). It was cut into small pieces. It was washed and dried. It was spread on a tray and put in the oven at 80°C until it become crispy and fragrant. The dried lemongrass was ground into fine powder and used for experiments.(Fig 2 & 3).

Fig2: Dried Lemongrass

Fig3: Powdered Lemongrass

2.2 Extraction of lemongrass oil:

Extraction of lemongrass was done using maceration extraction and soxhlet extraction methods.

i] Maceration method:

Materials: Lemongrass powder, distilled water, funnel, filter paper, Beaker weighing balance,

Method: 20 gm of lemongrass powder was weighed and added in a beaker. 200 ml of distilled water was added as a solvent into the beaker (with 1:10 ratio) for effective extraction. The mixture was thoroughly stirred. The mixture was boiled at 100^0C. The beaker was covered with petri plate and was allowed to settle down at room temperature (20-25^0C) for 24-72 hours. The mixture was filtered using filter paper into a new beaker. The procedure was repeated with fresh solvent if needed to maximize extraction. The obtained extract was stored in Amber vials at room temperature to prevent degradation of bioactive compounds(Onoriode, 2023).

ii]Soxhlet extraction:(using Distilled water)

The Soxhlet method is a continuous extraction technique used to extract bioactive compounds from solid materials using a suitable solvent. The process involves repeated cycles of solvent boiling, condensation, and percolation through the sample. The solvent dissolves the desired compounds, which accumulate in the extraction chamber. This method enhances extraction efficiency by using heat and continuous solvent recycling, making it ideal for extracting essential oils and phytochemicals from plant materials like lemongrass.

Materials: Soxhlet apparatus (round bottom flask, soxhelet extractor, condenser), lemongrass powder, distilled water, muslin cloth, beaker, weighing balance,

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Method:5 gm of lemongrass powder were weighed and wrapped in muslin cloth, which was then secured with a knot. The wrapped powder was placed into the Soxhlet thimble. 100 ml of distilled water were added into the round-bottom flask. The Soxhlet apparatus was then assembled with the condenser and heating mantle. The solvent in the flask was heated using the heating mantle or water bath. As the solvent vaporized, it traveled through the condenser, condensed back into liquid form, and dripped into the extractor chamber containing the lemongrass powder. The solvent continuously dissolved the bioactive compounds from the lemongrass powder and returned to the flask after passing through the siphon tube. The process was allowed to continue for 6-8 hours or until the solvent in the siphon tube became clear, indicating complete extraction. (Onoriode, 2023)

2.3 Qualitative Phytochemical Analysis-

Qualitative phytochemical analysis is based on specific chemical reactions that detect the presence of various bioactive compounds in plant extracts. Different reagents interact withspecific phytochemicals, leading to observable changes such as color formation, precipitation, or fluorescence. Common tests include:

1. Test for alkaloids- Mayer's test
Materials: Lemongrass oil (sample)**,** Mayer's reagent, Distilled water, Concentrated HCl, Test tubes, Pipettes, Beaker, Dropper
Method:2ml extract and 1ml of conc. HCL was added into the same test tube along with a few drops of Mayer's reagent and mixed together. The result for the same was observed later. (Sharma, 2020)

2.Test for phenols- (Ferric Chloride test)
Materials: Lemongrass oil (sample), 5% Ferric chloride solution ($FeCl_3$), Test tubes,Pipettes, Beaker, Dropper, Stirring rod
Method:1ml extract was added along with1ml 5% $FeCl_3$in a test tube. The result for the same was observed later.

3.Test for Flavonoids- (Lead Acetate test)
Materials: Lemongrass oil (sample), 10% Lead acetate solution, Distilled water, Test tubes, Pipettes, Beaker, Dropper, Stirring rod.
Method:1ml extract was added in test tube along with 1ml of 10% lead acetate solution .The result for the same was observed later.

4.Test for Tannins- (Braymer's Test)
Materials: Lemongrass oil (sample), 5%Ferric chloride, Distilled water, Test tubes, Pipettes, Beaker, Dropper, Stirring rod
Method:0.5 ml extract was added in the test tube.1ml distilled water and 1ml 5% ferric chloride were also added in the same test tube.The solution was stirred gently using a glass rod. The result for the same was observed later. (Sharma, 2020)

5.Test for Saponins- (Foam Test)
Materials: Lemongrass oil sample, Distilled water, Test tubes, Pipette, Glass rod
Method:1ml extract and 1ml D/W was added.The test tube was closed using a stopper. The test tube was vigorously shaken for about 30 seconds.The mixture was allowed to stand undisturbed for 10–15 minutes.The result for the same was observed later.

6.Test for Cardiac Glycosides- (Keller- Killiani Test)
Materials: Lemongrass oil sample, Distilled water, Glacial acetic acid, 5%Ferric chloride ($FeCl_3$) Concentrated sulfuric acid (H_2SO_4), Test tubes, Pipette, Glass rod
Method:5ml extract, 2ml of glacial acetic acid and 1ml of 5% $FeCl_3$ were added to a test tube.1 mL of concentrated sulfuric acid was added along the walls of the test tube using a pipette to form a separate layer at the bottom.Do not mix the solution after adding sulfuric acid.Observe the interface between the two layers.A reddish-brown ring at the interface, followed by a bluish-green coloration, indicates the presence of cardiac glycosides. (Sharma, 2020)

7.Test for Terpenoids- (ferric chloride test)
Materials: Lemongrass oil extract, 10% Ferric chloride, Test tubes, Pipette, Glass rod, Distilled water
Method:1ml extract was in the test tube.2ml of Distilled water was added in the same tube.In same test tube 1ml 10% $FeCl_3$ was added. Formation of intense color was observed.

8.Test for Quinones-
Materials: Lemongrass oil sample, Concentrated hydrochloric acid (HCl), Test tubes, Pipette, Glass rod
Method: Add 1ml extract in the test tube.In the same test tube add 0.5 ml of conc. HCL. Formation of yellow precipitate was observed.(Sharma, 2020)

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9.Test for Coumarins-
Materials: Lemongrass oil sample, 10% Sodium hydroxide (NaOH) solution, Test tubes, Pipette, Glass rod
Methods: Add 1 ml extract in test tube.In the same tube add 1.5 ml of 10% NaOH.Mix the solution gently and allow it to stand for a few minutes.The result for the same was observed later.

10.Test for Reducing Sugar
Materials: Lemongrass oil sample, Fehling's A solution ($CuSO_4$ solution), Fehling's B solution (Alkaline sodium potassium tartrate solution), Distilled water, Test tubes, Pipette, Beaker, Water bath, Glass rod
Method: Add 0.5 ml extract in the test tube.Add 1ml of D.W in the same tube.In a separate test tube, mix equal volumes (1 mL each) of Fehling's A and Fehling's B solutions.The mixture should appear deep blue due to the presence of Cu^{2+} ions.Add 2 mL of the prepared Fehling's reagent to the test tube containing the lemongrass oil-water mixture. Mix gently.Brick- red precipitation was observed. (Sharma, 2020)

11.Test for Glycoside
Materials: Lemongrass oil sample, Molisch's reagent (5% α-naphthol in ethanol), Concentrated sulfuric acid (H_2SO_4), Test tubes, Pipette, Distilled water, Water bath
Method: Add 2ml of extract in the test tube.Add 5ml of molisch's reagent in the same tube.After that add 1-2 drops of conc. H2SO4**.** Observe violet colour formation. (Sharma, 2020)

12.Test for Volatile Oils
Materials: Lemongrass oil sample, 10% Sodium hydroxide (NaOH) solution, Distilled water, Test tubes, Pipette,Glass rod
Methods: Add 2ml extract in the tube shake well to form an emulsion.Slowly add 2 mL of 10% NaOH solution to the mixture while stirring.Turbidity (cloudiness) or white precipitate formation was observed. (Sharma, 2020)

13.Test for Steroids-
Materials: Lemongrass oil sample**,** Glacial acetic acid**,** Concentrated sulfuric acid (H_2SO_4), Test tubes**,** Pipette**,** Glass rod
Methods: Add 2 mL of extract in the test tube. Add 2 mL of glacial acetic acid in the same test tube. Gently mix the solution by swirling. Carefully add 1 mL of concentrated sulfuric acid by pouring it down the side of the test tube to form a separate layer. Any color changeswas observed at the interface or within the solution (Sharma, 2020).

2.4 Anti-inflammatory Activity
Anti-inflammatory activity is assessed by evaluating the ability of a compound or extract to reduce inflammation-related responses, such as enzyme inhibition, membrane stabilization, or cytokine suppression. The principle is based on measuring the suppression of inflammatory mediators like prostaglandins, histamines, and cytokines using biological or chemical assays.
Proteinase inhibitory Assay-
The Proteinase Inhibitory Assay is used to evaluate the ability of a sample (e.g., plant extract) to inhibit proteolytic enzymes like trypsin or proteinase K. The principle is based on the enzymatic hydrolysis of protein substrates, which releases peptides or amino acids that can be measured spectrophotometrically.
Materials: Trypsin, Tris-HCL buffer (PH 7.4), Lemongrass oil extract (different conc.), 0.08% (w/v) Casein, 70% Perchloric acid, Distilled water, Falcon Tubes, Glass Piepette, Micropiepette, Microtips, Test tube stand, Incubator, Centrifuge, spectrophotometer

Method: The test samples (plant extracts) were dissolved in an appropriate solvent, such as distilled water, at different concentrations (25%, 50%, 75%, 100%). A stock solution of trypsin was prepared to a final concentration of 0.06 mg/mL. The reaction mixture, consisting of 2 mL, contained 1 mL extract, 1 mL Tris-HCl, and 0.06 mg trypsin. This mixture was placed in all the different concentration tubes. The reaction mixture was incubated at 37°C for 5 minutes. Then, 1 mL of 0.08% casein was added to all the tubes. The reaction mixture was incubated for an additional 20 minutes. After incubation, 2 mL of 70% perchloric acid was added to all the tubes to stop the reaction. The cloudy suspension was centrifuged at 5000 rpm for 15 minutes. The absorbance of the supernatant was measured at 280 nm against Tris-HCl buffer as a blank (Naz, 2017).

$$\textbf{Proteinase inhibition}(\%) = \frac{\textbf{Absorbance of Blank} - \textbf{Absorbance of test sample}}{\textbf{Absorbance of Blank}} \times \mathbf{100}$$

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Absorbance of Blank = Absorbance of the reaction mixture without the test sample.
Absorbance of Test Sample = Absorbance of the reaction mixture with the test sample

2.5 In Vitro Antibacterial Testing:
Minimum Inhibitory Concentration (MIC): Broth Dilution Method
MIC is the lowest concentration of the extract that inhibits visible growth of the bacterium.

Materials: Lemongrass extract, Media (Nutrient broth), Petri plates, micropipette, bumper tubes, ethanol, Distilled Water (control), bacterial culture, test tube stand.

Method:
1. Nutrient broth and bumper tubes were autoclaved at 1210C at 15 psi for 30 minutes.
2. Addition in the tubes was done according to Table 1.

Sr. No	Concentration of Lemongrass Extract (ml)	Amount of extract (ml)	Amount of Diluent (ml) (NB)	Total Volume (ml)
1	25%	0.75	4.25	5
2	50%	2.5	2.5	5
3	75%	3.75	1.25	5
4	100%	5	0	5

Table No.1: Broth Dilution method using Gram positive and Gram negative bacteria (E-coli) and (Bacillus subtilis).

3. Microorganisms (E-coli) and (Bacillus subtilis) were added to each tube in the quality of 0.1 ml.
4. The OD of the culture for addition was set at 0.1 at 540nm.
5. After addition all the tubes were incubated at 370^{C} for 24 hours. (Rabia Naz, 2017).

$$\textbf{Growth inhibition}(\%) = \frac{\textbf{Control OD} - \textbf{Treated OD}}{\textbf{Control OD}} \times \mathbf{100}$$

Control OD = Absorbance of the reaction mixture without the test sample.
Sample OD= Absorbance of the reaction mixture with the test sample.

2.6 Development of Incense Sticks
Materials- Charcoal, Neem powder, Starch, Marigold flower powder, Camphor, Sandalwood Powder, Lemongrass extract, Rose water, Bomboo sticks, Jumbo petri plate, Beaker.

Method-
1. Semi - solid paste of the given components was made (Table 2).

Component name	Grams
Charcoal	5 gm
Neem powder	1.25 gm
Starch	5 gm
Marigold flower powder	2.5 gm
Camphor	2.5 gm
Sandalwood Powder	1.25 gm
Lemongrass extract	Q.S
Rose water	Q.S

Table No.2: Components of Lemongrass Incense sticks. (**Ref)**

2. The paste was rolled into sticks using a binder like starch to hold the ingredients together.
3. The formed sticks were allowed to dry completely.
4. The incense sticks were tested for their fragrance and effectiveness (Pangavhane, 2024).

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III. RESULT AND DISCUSSION

3.1 Qualitative Phytochemical Analysis-

Qualitative phytochemical analysis is based on specific chemical reactions that detect the presence of various bioactive compounds in plant extracts. Different reagents interact with specific phytochemicals, leading to observable changes such as color formation, precipitation, or fluorescence.

Test Name	Predicted Result	Observed Result
Test for Alkaloids	White/ Green Color	-
Test for Phenols	Reddish Brown Color	-
Test for Flavonoids	Yellow Color	+
Test for Tannins	Blue- Green Color	-
Test for Saponins	Foam Formation	+
Test for Cardiac Glycosides	Brown ring/Violet ring	+
Test for Terpenoids	Intense color	+
Test for Quinones	Yellow Color	+
Test for Coumarins	Yellow Color	+
Test for Reducing Sugar	Brick Red Color	-
Test for Glycosides	Violet Color	+
Test for Volatile Oil	White Color	+
Test for Steroids	Blue- Green Color	-

Table No.3: Phytochemical Analysis of Lemongrass.

The results of the various phytochemical screening tests obtained during the experiment are shown in Table 7. Flavonoids, Saponins, Cardiac Glycosides, Terpenoids, Quinones,Coumarins, Glycosides, Volatiole oilsetc, were the phytoconstituents found in plants. According to the literature and the tally done with the obtained result, flavonoid caused risk reduction mainly from cardiovascular diseases and cancer. The presence of classes of phytochemical such as flavonoid showed cytotoxic effect. The color and aroma imparting flavonoids were stated to show anticancer properties. Additionally, cholesterol-lowering, as well as cytotoxic qualities, anti-bacterial, anti-viral properties, are credited to the presence of saponin. Plants containing a high amount of flavonoids could be useful as anti-bacteria. So the plants like lemon grass, Zingiber, Curcuma, and Acorus could be used as antibacterial, antiseptic agents. The plants containing phenolic compounds could be useful as an antioxidant. Quinine showed antipyretic property, so the plants containing quinine like Ocimum, Nyctanthes, Mentha, etc, could be used to reduce fever. The phenolic compound, tannin, terpenoid, and flavonoids possess an anti-helmintic property so the plant Zanthoxylum, Acorus could be used to treat stomach problems. The polyphenolic compounds, flavonoids, terpenoids found in Allium cepa, and Allium sativum are useful as an antioxidant, anti-inflammatory, and antibacterial agent. Likewise, they play an important role in reducing blood pressure, in preventing heart diseases. In conclusion, the findings of this study were aligned with the findings of Sharma, et al., 2020

3.2Proteinase inhibitory assay-

The Proteinase Inhibitory Assay is used to evaluate the ability of a sample (e.g., plant extract) to inhibit proteolytic enzymes like trypsin or proteinase K. The principle is based on the enzymatic hydrolysis of protein substrates, which releases peptides or amino acids that can be measured spectrophotometrically.

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Sample	Concentrations (%)	Proteinase inhibition (%)
Lemongrass oil (Maceration extract)	25 50 75 100	55.8 59.66 42.54 50
Lemongrass oil (Soxhlet extract)	25 50 75 100	30.93 27.9 30.11 31.21

Table No.4: Proteinase inhibitory assay

The proteinase inhibitory assay was conducted to assess the ability of lemongrass extracts (maceration and Soxhlet) to inhibit proteolytic enzymes. The results for both extraction methods are summarized in Table 8. In maceration extract, the highest inhibition was observed at 50% concentration (59.66%), while the lowest inhibition was at 75% concentration (42.54%). In soxhlet extract, the inhibition percentage remained lower compared to the maceration extract, with the highest inhibition at 100% concentration (31.21%) and the lowest at 50% concentration (27.9%). These findings indicate that the maceration extract demonstrated greater proteinase inhibitory activity across all concentrations compared to the Soxhlet extract. The difference in proteinase inhibition between the two extraction methods suggests that the maceration technique is more effective in preserving and extracting bioactive compounds responsible for enzyme inhibition. At higher concentrations (75% and 100%), the inhibitory activity of the maceration extract showed a decline (42.54% and 50%, respectively), which might be due to enzyme saturation or competitive inhibition mechanisms, where excess extract components interact with the enzyme in ways that reduce inhibition efficiency. Overall, these results indicate that maceration is the preferred extraction method for obtaining potent proteinase inhibitors from lemongrass. Further research should focus on identifying the specific bioactive compounds responsible for the inhibition and optimizing extraction conditions to maximize their yield and activity. In conclusion, the findings of this study were aligned with the findings of Naz, et al

3.3 Broth Dilution –

The broth dilution method is used to determine the minimum inhibitory concentration (MIC) of an antimicrobial agent against bacteria or fungi. The principle is based on serial dilution of the antimicrobial agent in a liquid growth medium (broth) followed by inoculation with a standardized microbial suspension.

Sample	Concentrations (%)	Percentage inhibition (%)	Percentage inhibition (%)
Lemongrass oil (Maceration extract)	25 50 75 100	33.33 - 46.66 36.66	Negative 17.14 40 51.42
Lemongrass oil (Soxhlet extract)	25 50 75 100	-	-

Table No.5: Broth Dilution Assay.

Key: - means no inhibition

The broth dilution method was used to determine the antimicrobial activity of lemongrass oil extracts (maceration and Soxhlet) against Escherichia coli (E. coli) and Bacillus subtilis. The minimum inhibitory concentration (MIC) was evaluated by measuring absorbance at 620 nm, and the percentage inhibition was calculated. The experimental findings (Table 9) show significant differences in antimicrobial activity between maceration and Soxhlet extracts. Maceration

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extract (E. coli). The highest inhibition (46.66%) was observed at 75% concentration, while at 50% concentration, the inhibition was negative, indicating bacterial growth stimulation. Maceration extract (Bacillus subtilis): The highest inhibition (51.42%) occurred at 100% concentration, showing strong antibacterial activity. Soxhlet extract (E. coli&Bacillus subtilis). The Soxhlet extract exhibited no inhibition across all concentrations, as indicated by negative values, meaning bacterial growth was not affected. The results demonstrate that maceration extraction is more effective in inhibiting bacterial growth than Soxhlet extraction. Maceration Extract showed moderate to strong inhibition of E. coli and Bacillus subtilis, particularly at higher concentrations (75% and 100%). The decreased inhibition at 25% and 50% concentrations for E. coli suggests that lower concentrations may not be sufficient to exert a strong antimicrobial effect. The highest inhibition (51.42% for Bacillus subtilis) at 100% concentration suggests that lemongrass oil contains potent antibacterial compounds like citral and limonene that disrupt bacterial cell membranes. In soxhlet extract, no inhibition was observed for both E. coli and Bacillus subtilis, suggesting that the Soxhlet method may have degraded key antimicrobial compounds due to prolonged heat exposure. Previous studies indicate that high temperatures can reduce the antimicrobial activity of essential oils, leading to lower efficacy in Soxhlet extracts (Ahmed et al., 2021). Maceration extraction is superior for preserving the antibacterial compounds in lemongrass oil. Soxhlet extraction was ineffective, likely due to the degradation of active compounds. Further studies are needed to optimize extraction conditions and identify the specific bioactive compounds responsible for antibacterial activity. The Results of this particular MIC tests were found not in line with the findings of Mohan et al., 2018

3.4 Development of Incense Sticks

The development of incense sticks is based on the principle of absorption, binding, and controlled combustionto release aromatic compounds when burned. A mixture of powdered combustible materials (e.g., wood powder, charcoal) and binding agents (e.g., jigat powder, gum) is used to form a uniform paste, which is then coated onto bamboo sticks. Essential oils, herbal extracts (e.g., lemongrass extract), or synthetic fragrances are added to enhance the aroma and provide additional benefits such as insect repellency or therapeutic effects. After drying, the sticks burn slowly and evenly, releasing fragrant smoke.

Fig 4. Incense Stick before binding

Fig 5. Incense Stick after binding.

Fumigation is useful to prevent disease by disinfection of the desired place. The inhibition rate observed was 99% which indicate that highly effective fumigation process in controlling microbial growth.

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This high rate of inhibition states that the fumigant is effective against the micro-organisms. This level of effectiveness is ideal for applications where it is critical to control microbial or fungal growth, such as in food preservation, agricultural storage, or medical sterilization.

Fig 6. Before Fumigation

Fig 7. After Fumigation.

The strong antimicrobial activity of lemongrass essential oil is attributed to its high content of citral, limonene, and myrcene, which have been reported to exhibit strong antibacterial, antifungal, and antiviral properties (Asif et al., 2020). The fumigation process works in the following ways:

Vapor Phase Diffusion: The active compounds in lemongrass incense sticks are released into the air, where they interact with microbial cells, disrupting their membranes and inhibiting their growth.

Oxidative Stress Induction: Lemongrass components generate reactive oxygen species (ROS), which cause oxidative damage to microbial DNA and proteins, leading to cell death (Gupta et al., 2019).

Disruption of Cellular Metabolism: Citral, a key component in lemongrass, interferes with enzyme activity and metabolic pathways in bacteria and fungi, rendering them inactive (Alara et al., 2019).

Lemongrass fumigation offers several advantages over traditional chemical fumigants like formaldehyde and methyl bromide, which are toxic and hazardous to human health. Unlike chemical fumigants, lemongrass:

Is natural and biodegradable, making it eco-friendly.Has no toxic residues, ensuring safety for food storage and medical applications. Provides pleasant aroma and acts as a natural air freshener while controlling microbial contamination.In conclusion, the findings of this study were aligned with the findings ofPangavhane, et al., 2024

IV. CONCLUSION-

The creation and assessment of herbal and natural incense sticks formulations for environmental purification is the main focus of the current work. The study successfully demonstrated the potential of lemongrass oil in incense stick formulation, antimicrobial efficacy, anti-inflammatory properties, and air purification through fumigation. The combination of biochemical testing, MIC analysis, and fumigation studies supports the application of lemongrass oil as a natural, eco-friendly alternative to synthetic fragrances and chemical disinfectants. The findings encourage further commercial development and large-scale production of lemongrass oil-based products for health, hygiene, and environmental benefits.The aforementioned findings make it clear that these incense sticks has the ability to disinfect a variety of surfaces and to purify the surrounding air. For disinfection purposes in hospitals and other facilities, this herbal incense sticks with a defined quality that is made from easily accessible and reasonably priced sources can be used in place of chemical sources and harmful UV rays. The current work can be expanded to include public latrines, hospitals, colleges, and schools, among other places. It can serve as an air freshener and room purifier in addition to aiding in the creation of a positive atmosphere.

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