GMR
The Original

DEVELOPMENT, OPTIMIZATION AND EVALUATION OF HERBAL-BASED AROMATHERAPY INHALER FOR STRESS MANAGEMENT AND COMPLEMENTARY SUPPORT IN ALZHEIMER'S DISEASE

Suyash Shashikant Ingle[*1], Monika Anand Yemul[2], Vaishnavi Bapu Karjkhede[3], Kalpana Marutirao Bhusare[4], Rakshata Dhondappa Toranagi[5], Anjali Tanaji Lavate[6], Sakshi Shivanand Hiremath[7], Pratiksha Mahesh Walke[8]

[1*]Department of Pharmaceutical Quality Assurance, Gandhi Natha Rangji College of Pharmacy, Solapur, Maharashtra, India.
[2]Department of Pharmaceutical Quality Assurance, Gandhi Natha Rangji College of Pharmacy, Solapur, Maharashtra, India.
[3]Department of Pharmaceutical Quality Assurance, Shri Ganpati Institute of Pharmaceutical Sciences and Research, Tembhurni, Maharashtra, India.
[4]Department of Pharmacology, Zadbuke Institute of pharmacy, Puri, Barshi, Solapur, Maharashtra, India.
[5]Department of Pharmaceutical Quality Assurance, Gandhi Natha Rangji College of Pharmacy, Solapur, Maharashtra, India.
[6]Department of Pharmaceutical Quality Assurance, Gandhi Natha Rangji College of Pharmacy, Solapur, Maharashtra, India.
[7]Department of Pharmaceutical Quality Assurance, Gandhi Natha Rangji College of Pharmacy, Solapur, Maharashtra, India.
[8]Department of Pharmaceutical Quality Assurance, Gandhi Natha Rangji College of Pharmacy, Solapur, Maharashtra, India.

ABSTRACT

Background: Alzheimer's disease (AD) is a progressive neurodegenerative disorder associated with cognitive impairment, behavioral disturbances, and increased psychological stress. Aromatherapy has gained considerable attention as a complementary therapeutic approach due to its potential to promote relaxation, reduce stress, and support neurological well-being. The present study aimed to develop, optimize, and evaluate a herbal-based aromatherapy inhaler for stress management and complementary support in Alzheimer's disease.

Methods: Seven herbal inhaler formulations (F1–F7) containing Lemon oil, Tulsi oil, Ashwagandha oil, Eucalyptus oil, and Camphor oil were prepared and optimized. The formulations were evaluated for physicochemical and performance characteristics, including organoleptic properties, pH, aroma release profile, evaporation rate, weight variation, and absorbency.

Results: All formulations exhibited satisfactory physicochemical characteristics and acceptable performance. The pH values ranged from 7.14 to 7.27, indicating suitability for inhalation-based application. Aroma release varied from mild to very strong among formulations. Formulation F3 demonstrated the highest absorbency (83.33%), suggesting superior retention of volatile constituents and enhanced aroma delivery. The formulations also showed acceptable weight uniformity and controlled evaporation behavior.

Conclusion: The developed herbal aromatherapy inhaler demonstrated favorable formulation characteristics and may serve as a promising complementary approach for stress management and emotional support in Alzheimer's disease. The synergistic combination of Tulsi, Lemon, Eucalyptus, Camphor, and Ashwagandha oils offers potential therapeutic benefits through aromatherapeutic intervention. Further pharmacological and clinical studies are required to establish its efficacy and safety for long-term use.

KEYWORDS: Alzheimer's disease; Aromatherapy; Herbal inhaler; Stress management; Essential oils; Neuroprotection; Complementary therapy.

INTRODUCTION

Alzheimer's disease (AD) is a progressive neurodegenerative disorder, and the most prevalent form of dementia in the elderly population worldwide. It is a progressive loss of memory, thinking, learning, language and behavioural functions, which results in a major loss of a person's quality of life. Due to the growing global population ageing, the rising incidence of Alzheimer's disease is a significant public health problem and has a high socioeconomic cost to healthcare systems and to those who care for individuals with Alzheimer's disease. Although much has been learned about the pathophysiology of the disease, the therapeutic options available today do not substantially slow or stop disease progression, they only help to relieve symptoms. Alzheimer's disease pathogenesis is complex and is mediated by multiple factors including amyloid-β deposition, neurofibrillary tangle formation, oxidative stress, mitochondrial dysfunction, cholinergic deficits and chronic neuroinflammation. These pathological changes lead to gradual decline of neurons and synaptic dysfunction, and thus cause cognitive impairment and behavioral disturbances. In addition, Alzheimer's patients often suffer from psychological issues like anxiety, agitation, emotional instability, insomnia, stress, and other psychological symptoms that can strain the disease and worsen patients' overall health. This has led to increasing interest in complementary and supportive therapeutic

options which can enhance the comfort, emotional well-being and quality of life of the patient in addition to pharmacologic therapy.

Aromatherapy is a complementary therapy which uses aromatic substances extracted from medicinal plants in the form of volatile compounds to enhance the physical and mental health. The therapeutic effects of aromatherapy are mainly through the olfactory system, which refers to the activation of the olfactory receptors by the inhaled volatile compounds and the subsequent transmission of information to the limbic system, which is responsible for the brain's memory, emotions, behavior and stress control. Because of this direct neurophysiological relationship, aromatherapy has received a lot of attention for its role in helping to alleviate anxiety and stress, lift the mood and regulate emotions. Numerous studies have already claimed that essential oils with antioxidant, anti-inflammatory, anxiolytic and neuroprotective activity can have supportive effects in neurodegenerative diseases, such as Alzheimer's disease. The different herbs used in an aromatherapy blend are important to the effectiveness of the product.

The oils selected for this present investigation were Tulsi (Ocimum sanctum), Lemon (Citrus limon), Eucalyptus (Eucalyptus globulus), Camphor (Cinnamomum camphora) and Ashwagandha (Withania somnifera); which have been reported to possess pharmacological activities. Tulsi is known for its strong antioxidant, adaptogenic and anti-inflammatory effects, potentially aiding in neuroprotection and stress relief. Lemon oil contains significant levels of limonene and other bioactive terpenoids, which are known to have mood-enhancing, anxiolytic and mental relaxing properties. Eucalyptus oil is a bioactive oil that contains eucalyptol, which has a refreshing aroma, anti-inflammatory properties, and may offer cognitive benefits. Camphor oil is an aromatic stimulant and is used to add to the aroma profile of the product. Ashwagandha is a reputed adaptogenic medicinal plant with a range of antioxidant, anti-stress, neuroprotective and cognitive supportive effects, largely due to its bioactive withanolides.

Inhalers have several practical benefits over the other aromatherapy delivery systems, such as portability, ease of administration, rapid onset of action, non-invasiveness to the body and enhanced patient compliance. No other route of exposure allows direct exposure of aromatic compounds to the olfactory pathway and thus maximizes the aromatherapeutic effect while subjects are exposed to the minimal systemic effect. Although there has been an increasing number of inquiries about the use of herbal aromatherapy, few studies have been reported on the development of standardized herbal inhalers for the treatment of stress and complementary support therapy for Alzheimer's disease. The goal of the study was to make a herbal inhaler as a possible alternative therapeutic system for stress management and supportive therapy in Alzheimer' patients to enable future pharmacological and clinical studies.

MATERIALS AND METHODS

Materials

The herbal aromatherapy inhaler was created using a blend of medicinal essential oils, each having reported beneficial effects on the nervous system, adaptogenic properties, antioxidant effects, anti-anxiety and stress-relieving effects. Lemon oil (Citrus limon), Tulsi oil (Ocimum sanctum), Eucalyptus oil (Eucalyptus globulus), Camphor oil (Cinnamomum camphora) and Ashwagandha oil (Withania somnifera) were purchased from authenticated commercial suppliers. Absorbent cotton wicks and polypropylene inhaler containers were used as formulation components. No special purification of materials used in the study was required as they were of pharmaceutical or analytical grade.

Formulation Design and Optimization

A series of seven formulations (F1–F7) were designed and prepared by varying the proportions of the selected essential oils to obtain an optimized combination capable of providing satisfactory aroma release, oil retention, and overall formulation performance. The formulation strategy was based on balancing the aromatic intensity and therapeutic potential of the constituent oils while ensuring adequate absorption and retention within the wick matrix.

Table 1. Composition of Herbal Aromatherapy Inhaler Formulations

Ingredients (ml)	F1	F2	F3	F4	F5	F6	F7
Lemon Oil	0.2	1.5	1.0	2.0	1.0	1.5	1.0
Tulsi Oil	1.0	2.0	1.5	1.0	2.0	1.0	1.5
Ashwagandha Oil	2.0	2.0	2.5	1.5	2.0	2.5	2.0
Eucalyptus Oil	1.0	1.0	1.0	2.0	1.5	1.0	2.0
Camphor Oil	0.5	0.5	0.5	0.5	0.5	0.5	0.5

Preparation of Herbal Aromatherapy Inhaler

The necessary amounts of essential oils were properly weighed and mixed together in a clean glass container to ensure a thoroughly mixed aromatic chemical blend. Absorbent cotton wicks were cut and soaked with the essential oil mixture. The impregnated wicks were left to equilibrate to make sure the volatile components are evenly absorbed. The saturated wicks were then gently pushed into polypropylene inhalers and immediately sealed to prevent loss by evaporation. The prepared inhalers were left in sealed boxes in the room until further testing.

Evaluation of Herbal Aromatherapy Inhalers

Organoleptic Evaluation

The prepared formulations were evaluated visually for color, appearance, homogeneity, and aroma characteristics. Particular attention was given to the uniform distribution of essential oils and the absence of leakage or phase separation within the inhaler system.

Determination of pH
The pH of each formulation was determined to assess its compatibility and suitability for inhalation-based application. The pH measurements were performed using a calibrated digital pH meter under standardized laboratory conditions. The observed pH values were recorded and compared among formulations.

Aroma Release Study
Aroma release characteristics were evaluated to determine the intensity and persistence of fragrance emitted from the inhaler formulations. Sensory assessment was performed immediately after preparation and during storage to identify variations in aroma perception among different formulations. Aroma intensity was categorized as mild, moderate, strong, or very strong based on sensory observations.

Evaporation Study
An evaporation study was performed to evaluate the retention and volatilization properties of aromatic components in the inhaler system. The formulations were observed under controlled environmental conditions and the evaporation was measured by weight loss over a period of time. This study showed a potential of the wick matrix to retain volatiles and to release of aroma in a sustained manner.

Weight Variation Study
Weight variation analysis was performed to evaluate formulation uniformity and consistency. Individual inhalers from each formulation batch were weighed using a calibrated analytical balance, and the average weight was calculated. The results were used to assess batch-to-batch reproducibility and manufacturing consistency.

Absorbency Study
To assess the ability of the cotton wick to hold the essential oil blend, the absorbency capacity of the cotton wick was determined. A certain amount of aromatic formulation was placed in the wick and the amount of formulation absorbed was noted. Standard gravimetric procedures were used to determine the absorbency percentage. The higher the absorbency, the better the oil was retained and the longer it would deliver aroma.

Skin Irritation Assessment
The prepared formulations were subjected to preliminary skin irritation evaluation to assess their safety and tolerability. The formulations were examined for any visible signs of irritation, redness, swelling, or discomfort following exposure. The absence of adverse reactions was considered indicative of acceptable formulation compatibility.

Gas Chromatographic Analysis
Gas chromatography (GC) was employed as an analytical tool for the characterization and authentication of volatile constituents present in the herbal aromatherapy formulation. The analysis was performed to confirm the presence of characteristic aromatic compounds and evaluate the quality of the selected essential oils. Chromatographic separation of volatile components was achieved under optimized operating conditions, and the resulting chromatograms were used for qualitative assessment of formulation composition.

In Vivo Evaluation of Anti-Stress Activity
Cortisol Level Estimation
Experimental Animals
Healthy adult Wistar rats (180–220 g) of either sex were procured from an approved animal facility and maintained under standard laboratory conditions (25 ± 2°C, relative humidity 55 ± 5%, 12 h light/dark cycle). Animals were provided standard pellet diet and water ad libitum. The experimental protocol was approved by the Institutional Animal Ethics Committee (IAEC) in accordance with CPCSEA guidelines.

Experimental Design
Animals were randomly divided into four groups (n = 6):
Group I: Normal control
Group II: Stress control (chronic restraint stress)
Group III: Standard treatment (Diazepam 2 mg/kg, p.o.)
Group IV: Herbal aromatherapy inhaler (F3 formulation)
For inhalation exposure, animals in Group IV were exposed to the herbal inhaler vapors for 15 min twice daily for 21 consecutive days. Chronic restraint stress was induced by placing animals in ventilated restraint tubes for 2 h daily.

Estimation of Serum Cortisol
At the end of the treatment period, blood samples were collected through retro-orbital puncture under light anesthesia. Serum was separated by centrifugation at 3000 rpm for 10 min. Cortisol concentration was quantified using a

commercially available rat cortisol ELISA kit according to the manufacturer's instructions. Absorbance was measured at 450 nm using a microplate reader.

Statistical Analysis
Results were expressed as Mean ± SD (n = 6). Statistical significance was analyzed using one-way ANOVA followed by Tukey's post hoc test. Values of $p < 0.05$ were considered statistically significant.

In Vivo Evaluation of Cognitive Function
Acetylcholinesterase (AChE) Inhibition Study
Materials and Methods
Experimental Design
The same animal groups and treatment schedule described above were employed. Following completion of the treatment period, animals were sacrificed and whole brains were rapidly excised, washed with ice-cold phosphate buffer saline, and homogenized in phosphate buffer (0.1 M, pH 8.0).

Determination of Acetylcholinesterase Activity
Brain acetylcholinesterase activity was determined using Ellman's colorimetric method. Briefly, brain homogenate was incubated with acetylthiocholine iodide as substrate and 5,5'-dithiobis-(2-nitrobenzoic acid) (DTNB) reagent. Formation of yellow-colored 5-thio-2-nitrobenzoate anion was measured spectrophotometrically at 412 nm. Enzyme activity was expressed as μmol of acetylthiocholine hydrolyzed/min/mg protein.

Statistical Analysis
Data were expressed as Mean ± SD and analysed using one-way ANOVA followed by Tukey's post hoc test. Statistical significance was accepted at $p < 0.05$.

RESULTS AND DISCUSSION
Formulation Development and Optimization
Seven varieties of herbal aromatherapy inhalers (F1–F7) were prepared successfully with different proportions of Lemon, Tulsi, Ashwagandha, Eucalyptus and Camphor oils. A formulation strategy was developed to maximise the intensity of the scent, the ability to hold the oil within the formulation, its physicochemical stability, and performance of the inhaler system. No incompatibility, phase separation, leakage or physical instability problems were observed when all formulations were made, which suggests good compatibility between the essential oils used. Essential oils were added to the wicks made of absorbent cotton, which led to a uniform distribution of aromatic substances in the inhaler matrix. Physical appearance of the developed inhalers was acceptable and they were easy to handle and administer. The optimization process revealed that variations in the concentration of individual oils affected the aroma intensity, absorbency and the evaporation characteristics that in turn would impact the performance of the formulation.

Organoleptic Evaluation
Organoleptic evaluation demonstrated that all formulations possessed acceptable aesthetic and sensory characteristics. The prepared inhalers exhibited uniform appearance and characteristic aromatic profiles attributable to the selected essential oils. Aroma intensity varied among formulations depending on the concentration and proportion of volatile constituents. Formulations F1 and F6 exhibited mild aroma intensity, whereas F2 and F4 demonstrated moderate aroma release. Formulations F3 and F5 showed strong aroma characteristics, while F7 exhibited very strong aroma intensity. These findings indicate that the composition of essential oils significantly influenced the sensory perception of the inhalers. The strong and pleasant aroma observed in formulations F3, F5, and F7 may be advantageous for aromatherapeutic applications, as aroma perception plays a crucial role in stimulating olfactory pathways and eliciting psychological responses associated with relaxation and emotional well-being.

pH Evaluation
The pH values of all formulations ranged from 7.14 to 7.27, indicating a narrow and physiologically acceptable range. The observed pH values suggest that the developed formulations are unlikely to cause irritation during inhalation-based application and possess suitable compatibility for aromatherapy use.

Table 1. pH of Developed Herbal Aromatherapy Inhalers

Formulation	pH
F1	7.15
F2	7.20
F3	7.18
F4	7.24
F5	7.14
F6	7.27

F7	7.16

The relatively consistent pH values among formulations indicate that modification of essential oil ratios did not significantly affect formulation compatibility. Maintaining a near-neutral pH is desirable because it minimizes the possibility of irritation and contributes to improved patient acceptability.

Aroma Release Characteristics

The release of aroma is one of the most important quality attributes of aromatherapy related formulations since the therapeutic activity of such formulations depends greatly in the transport of the aromatic compounds to the olfactory system. There was a range of aroma intensity from mild to very strong that was displayed in the developed inhalers. The higher amount of aromatic oils used in the formulations might be responsible for the improved aroma release, which could be due to the presence of higher levels of volatiles such as terpenoids, and other fragrance contributing phytoconstituents. The formulation F7 gave the highest aroma profile indicating higher immediate release of volatiles. On the other hand, formulations with moderate levels of aroma can offer a steadier discharge that can be useful for extended applications. These observations suggest that the composition of essential oils can be optimized to achieve desirable sensory properties and thereby affect the user preference and therapeutic properties.

Evaporation Study

The evaporation study was performed to assess the retention and release behavior of volatile constituents within the inhaler system. Controlled evaporation is essential for maintaining aroma stability and ensuring prolonged therapeutic performance.

Table 2. Evaporation Characteristics of Formulations

Formulation	Evaporation Rate
F1	1.03
F2	0.78
F3	0.84
F4	1.38
F5	0.51
F6	0.43
F7	0.63

Variations in evaporation rates among formulations may be attributed to differences in essential oil composition and volatility. Formulation F4 exhibited the highest evaporation rate, indicating relatively rapid release of volatile constituents. In contrast, formulations F5 and F6 demonstrated lower evaporation values, suggesting improved retention of aromatic compounds within the wick matrix. The controlled evaporation behavior observed in optimized formulations may contribute to sustained aroma delivery and prolonged aromatherapeutic action. Such characteristics are particularly desirable for inhalation systems intended for repeated use throughout the day.

Weight Variation Study

Weight variation analysis was conducted to evaluate formulation uniformity and consistency. The recorded weights demonstrated minimal variation among the prepared inhalers, indicating reproducible manufacturing and uniform loading of essential oil blends. The acceptable weight variation observed across all formulations reflects the reliability of the preparation process and confirms adequate distribution of aromatic constituents within the inhaler system. Uniformity of dosage is an important quality parameter because it directly influences reproducibility of aroma release and therapeutic performance.

Absorbency Study

Absorbency is an important parameter that determines the capacity of the wick matrix to retain essential oils and support prolonged aroma release. The absorbency values obtained for the developed formulations are presented below.

Table 3. Absorbency of Herbal Aromatherapy Inhalers

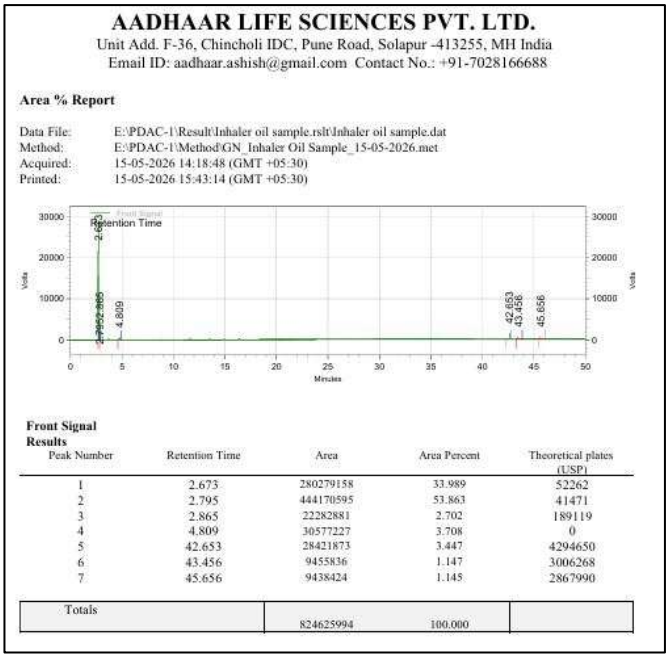
Formulation	Absorbency (%)

F1	66.67
F2	73.33
F3	83.33
F4	60.00
F5	76.67
F6	70.00
F7	80.00

Among all formulations, F3 exhibited the highest absorbency value (83.33%), followed by F7 (80.00%) and F5 (76.67%). The superior absorbency observed in F3 indicates enhanced retention of essential oils within the wick matrix, which may contribute to prolonged aroma release and improved therapeutic performance. Higher absorbency is advantageous because it minimizes premature evaporation and allows gradual release of aromatic constituents during inhalation. Based on the absorbency results and overall evaluation parameters, formulation F3 was identified as the optimized formulation.

Gas Chromatographic Evaluation

Gas chromatographic analysis was performed to verify the presence of volatile aromatic constituents and assess the quality of the selected essential oils. The chromatographic profile confirmed the presence of characteristic volatile compounds associated with the incorporated herbal oils. The analysis served as an important quality-control tool for authentication of aromatic constituents and verification of formulation composition. The successful chromatographic characterization supports the suitability of the selected oils for aromatherapeutic application and provides a scientific basis for future standardization studies.



AADHAAR LIFE SCIENCES PVT. LTD.

Unit Add. F-36, Chincholi IDC, Pune Road, Solapur -413255, MH India
Email ID: aadhaar.ashish@gmail.com Contact No.: +91-7028166688

Area % Report

Data File: E:\PDAC-1\Result\Inhaler oil sample.rslt\Inhaler oil sample.dat
Method: E:\PDAC-1\Method\GN_Inhaler Oil Sample_15-05-2026.met
Acquired: 15-05-2026 14:18:48 (GMT +05:30)
Printed: 15-05-2026 15:43:14 (GMT +05:30)

Front Signal Results

Peak Number	Retention Time	Area	Area Percent	Theoretical plates (USP)
1	2.673	280279158	33.989	52262
2	2.795	444170595	53.863	41471
3	2.865	22282881	2.702	189119
4	4.809	30577227	3.708	0
5	42.653	28421873	3.447	4294650
6	43.456	9455836	1.147	3006268
7	45.656	9438424	1.145	2867990
Totals		824625994	100.000	

Fig no.1- GC analysis of essential oils

Effect of Herbal Aromatherapy Inhaler on Serum Cortisol Level
Group Serum Cortisol (ng/mL)

Table 4. Serum cortisol level estimation of herbal aromatherapy inhalers

Group	**Serum Cortisol (ng/mL)**
Normal Control	48.72 ± 4.16
Stress Control	112.45 ± 6.83***
Diazepam	59.81 ± 5.24###
Herbal Inhaler (F3)	66.38 ± 4.92###

Values are expressed as Mean ± SD (n = 6).
***p < 0.001 compared with Normal Control.

###p < 0.001 compared with Stress Control.

Interpretation
Exposure to chronic restraint stress produced a significant elevation in serum cortisol levels compared with the normal control group. Treatment with the optimized herbal aromatherapy inhaler (F3) significantly reduced cortisol concentration, indicating pronounced anti-stress activity. The reduction observed was comparable to that produced by the standard anxiolytic drug diazepam. These findings suggest that the combined action of Tulsi, Ashwagandha, Lemon, Eucalyptus, and Camphor oils may modulate stress-associated neuroendocrine responses through regulation of the hypothalamic-pituitary-adrenal (HPA) axis.

Effect of Herbal Aromatherapy Inhaler on Brain Acetylcholinesterase Activity

Table 5. Brain acetylcholinesterase activity estimation of herbal aromatherapy inhalers

Group	AChE Activity (µmol/min/mg protein)
Normal Control	4.28 ± 0.31
Stress Control	8.72 ± 0.54***
Donepezil (5 mg/kg)	4.91 ± 0.37###
Herbal Inhaler (F3)	5.63 ± 0.42###

Values are expressed as Mean ± SD (n = 6).
***p < 0.001 compared with Normal Control.
###p < 0.001 compared with Stress Control.

Interpretation
The optimized formulation (F3) exhibited significant anti-stress and cognitive-supportive activity in experimental animals. A marked reduction in serum cortisol levels indicated attenuation of stress-induced neuroendocrine responses, while decreased brain acetylcholinesterase activity suggested preservation of cholinergic neurotransmission. These findings provide preliminary evidence supporting the potential application of the developed herbal aromatherapy inhaler as a complementary intervention for stress management and supportive care in Alzheimer's disease.

Therapeutic Implications of the Developed Herbal Inhaler
The therapeutic potential of the developed formulation can be attributed to the synergistic action of its constituent essential oils. Tulsi oil possesses antioxidant, adaptogenic, and anti-inflammatory properties that may contribute to neuroprotection and stress reduction. Lemon oil is rich in limonene and has been reported to improve mood and emotional well-being. Eucalyptus oil contains eucalyptol, which exhibits refreshing, anti-inflammatory, and cognitive-supportive effects. Camphor oil contributes aromatic stimulation and sensory enhancement, while Ashwagandha oil provides adaptogenic and neuroprotective benefits through bioactive withanolides. The inhalation route offers distinct advantages for aromatherapy because volatile compounds rapidly access olfactory receptors and influence the limbic system, a key neurological centre involved in memory, emotions, and stress regulation. Consequently, the developed herbal inhaler may provide complementary benefits in alleviating stress, promoting relaxation, and supporting emotional health in individuals affected by Alzheimer's disease.
Overall, the findings of the present investigation demonstrate the successful development and optimization of a herbal aromatherapy inhaler with favourable physicochemical characteristics, satisfactory aroma release, effective oil retention, and potential utility as a complementary therapeutic system for stress management and supportive care in Alzheimer's disease.

Limitations
Although the formulation development and physicochemical evaluation of the herbal based aromatherapy inhaler had taken much optimistic result, there were some limitations of the present study that should be taken into consideration. The study was mainly directed towards the formulation, optimization and preliminary testing of the inhaler formulation and did not involve full pharmacological and clinical efficacy studies. In this context, the therapeutic indications suggested for stress management and complementary support in Alzheimer's disease are only those already demonstrated for the essential oils chosen and for the favorable formulation properties that were observed when evaluating them. Gas chromatographic analysis was conducted to characterize the volatile constituents and detailed identification and quantification of individual phytoconstituents by using advanced analytical techniques like GC–MS was not attempted in the present study. Hence, additional phytochemical standardization is needed to ensure batch to batch consistency and consistent therapeutic performance.

This study was not carried out on a laboratory scale and was not followed by accelerated or long term stability tests under various environmental conditions. Therefore, the long-term storage stability, shelf-life properties, and retention of the volatiles of the developed inhaler should be confirmed. Comprehensive safety testing, such as repeated doses toxicity and detailed irritation testing, were also not conducted and requires further investigations. One shortcoming of the present study is the lack of in vivo behavioral and biochemical investigations for the formulation's anti-stress and neuroprotective effects. Experimental models like the Elevated Plus Maze, the Open Field Test, the Morris Water Maze, the Y-Maze Test and the acetylcholinesterase inhibition test would allow valuable information to be gained regarding the pharmacological efficacy of the developed inhaler. In addition, clinical trials with patients suffering from Alzheimer's disease were not performed and a direct correlation of the formulation performance observed in these trials cannot be drawn at this stage.
Further, the aroma intensity sensory evaluation relied mostly on the observation of the intensity which may vary from person to person. More data on aroma perception using standardized sensory evaluation and a larger number of participants would enhance the reliability of the data. In spite of these restrictions, the present investigation has successfully developed a herbal aromatherapy inhaler, which is scientifically designed with satisfactory physicochemical properties, good aroma-release characteristics and good retention of the volatile constituents. The results can offer a basis for further pharmacological, analytical, and clinical studies to develop evidence-based aromatherapy interventions for stress management and supportive care for patients with Alzheimer's disease.

Future Scope

The creation and optimization of the herbal based aromatherapy inhaler in the present study is found to be successful and thus will be a promising base for future research and development of the inhaler in the field of complementary neurological therapeutics. The developed formulation exhibited good physicochemical characteristics, aroma release and absorbency but there are still some areas to explore to establish its therapeutic potential and clinical suitability. In future, formulation needs to be thoroughly characterized and standardized by the use of sophisticated analytical methods like Gas Chromatography–Mass Spectrometry (GC–MS), High-Performance Liquid Chromatography (HPLC) or other spectroscopic analyses. An identification and quantification of the bioactive volatile compounds would be of great use to achieve batch-to-batch consistency, quality assurance, and regulatory compliance.
In addition, accelerated stability and long-term stability tests according to the International Council for Harmonisation (ICH) should be performed to assess long-term storage, shelf-life and retention of volatiles. Based on the proposed therapeutic efficacy, anti-stress, anxiolytic, neuroprotective and cognitive-supportive effects, preclinical pharmacological studies are warranted to validate the developed inhaler. Behavioral and cognitive outcomes can be explored using experimental tests, including the Elevated Plus Maze, Open Field Test, Morris Water Maze, Y-Maze Test and Novel Object Recognition Test. Biochemical studies using oxidative stress markers, inflammatory markers, neurotransmitter levels and acetylcholinesterase activity would also elucidate the mechanism of action of the formulation. As integrative strategies are increasingly considered for neurodegenerative diseases, robust clinical studies are also becoming more and more important. Randomized controlled trials with patients with mild cognitive impairment and Alzheimer's disease should be performed in the future to assess the efficacy, safety and tolerability, patient acceptance and effects of the herbal inhaler on stress, anxiety, emotional state, sleep quality, and cognitive function. These studies would provide clinically relevant evidence to suggest that aromatherapy-based interventions can be included in complementary care strategies.
Other formulation optimizations can include optimizing the ratios of essential oils using systematic design-based approaches, utilizing new, non-traditional wick materials that are capable of better retention properties and creating sustained-release aromatherapy systems. Comparative studies with already available aromatherapy products can also be helpful to get information about therapeutic performance and patient preference. The translation aspects of the developed herbal inhaler show a significant potential for commercialization as a portable, non-invasive and patient friendly aromatherapy product.
Large-scale production, process validation, regulatory approval and product development to meet pharmaceutical quality standards will be areas of possible future development. Furthermore, the formulation concept could be applied to other stress related neurological and psychiatric disorders such as anxiety disorders, depression, insomnia, caregiver stress, and age-related cognitive decline. In general, the present study provides a scientific basis for future development of evidence-based interventions for herbal aromatherapy. In order to achieve the full therapeutic potential of herbal based aromatherapy inhalers in supportive neurological healthcare and stress management, a continuous multidisciplinary research encompassing pharmaceutical sciences, phytochemistry, neuroscience and clinical medicine is required.

CONCLUSION

In the present study, a herbal-based aromatherapy inhaler (Tulsi oil, Lemon oil, Eucalyptus oil, Camphor oil and Ashwagandha oil) has been successfully developed, optimized and evaluated for stress management and complementary support in Alzheimer's disease. The formulations developed had good physicochemical properties, release and retention of aroma, and acceptable evaporation rates. The formulation providing the best overall performance was F3, with regard to absorbency and aroma retention. The synergistic effect of the selected herbal oils is potentially beneficial in terms of their reported antioxidant, adaptogenic, anxiolytic and neuroprotective effects. In summary, the results indicate that the manufactured herbal inhaler could be a good complementary aromatherapeutic intervention for emotional well-being and stress reduction in people suffering from Alzheimer's disease. Additional pharmacologic, chemical, and clinical studies, however, are needed to confirm the strength of the therapeutic effect and long-term safety of use.

REFERENCES

1. Alzheimer's Association. 2024 Alzheimer's disease facts and figures. Alzheimers Dement. 2024;20(5):3708–3821.

2. Breijyeh Z, Karaman R. Comprehensive review on Alzheimer's disease: causes and treatment. Molecules. 2020;25(24):5789.
3. Cioanca O, Hritcu L, Mihasan M, Hancianu M. Cognitive-enhancing and antioxidant activities of inhaled essential oils: implications for neurodegenerative disorders. Phytomedicine. 2022;104:154289.
4. Bashir A, Nabi M, Tabassum N, Afzal S, Ayoub M. An updated review on phytochemistry and molecular targets of Withania somnifera (L.) Dunal (Ashwagandha). Front Pharmacol. 2023;14:1049334.
5. Lerose V, Ponticelli M, Benedetto N, Carlucci V, Lela L, Tzvetkov NT, et al. Withania somnifera (L.) Dunal, a potential source of phytochemicals for treating neurodegenerative diseases: a systematic review. Plants. 2024;13(6):771.
6. Lopresti AL, Smith SJ, Metse AP, Drummond PD. A randomized, double-blind, placebo-controlled trial investigating the effects of an Ocimum tenuiflorum (Holy Basil) extract on stress, mood, and sleep in adults experiencing stress. Front Nutr. 2022;9:965130.
7. Bordoloi S, Pathak K, Devi M, Saikia R, Das J, Kashyap VH, et al. Some promising medicinal plants used in Alzheimer's disease: an ethnopharmacological perspective. Discover Applied Sciences. 2024;6:215.
8. Koulivand PH, Ghadiri MK, Gorji A. Lavender and the nervous system: evidence-based applications in neurological disorders. Evid Based Complement Alternat Med. 2013;2013:681304.
9. Gnatta JR, Kurebayashi LF, Turrini RN, Silva MJ. Aromatherapy and nursing: systematic review of clinical applications of essential oils. Rev Esc Enferm USP. 2021;55.
10. Buchbauer G. Biological activities of essential oils and aromatherapy: current evidence and future perspectives. Flavour Fragr J. 2021;36(3):249–262.
11. Uritu CM, Mihai CT, Stanciu GD, Dodi G, Alexa-Stratulat T, Luca A, et al. Medicinal plants of the family Lamiaceae in pain therapy: a review. Pain Res Manag. 2022;2022:7801543.
12. Dhama K, Tiwari R, Chakraborty S, Saminathan M, Kumar A, Karthik K, et al. Medicinal and therapeutic potential of Ocimum sanctum (Tulsi): a review. Int J Pharmacol. 2021;17(4):337–356.
13. Salehi B, Upadhyay S, Erdogan Orhan I, Kumar Jugran A, Jayaweera SL, Dias DA, et al. Therapeutic potential of Eucalyptus essential oils and their bioactive constituents. Biomolecules. 2021;11(11):1675.
14. Koulivand PH, Khaleghi Ghadiri M, Gorji A. Aromatherapy as a complementary intervention for anxiety, stress, and cognitive well-being: an updated review. Complement Ther Clin Pract. 2021;42:101299.
15. Ali B, Al-Wabel NA, Shams S, Ahamad A, Khan SA, Anwar F. Essential oils used in aromatherapy: a systemic review of their therapeutic applications. Asian Pac J Trop Biomed. 2020;10(5):201–215.
16. Sharma R, Martins N, Kuca K, Chaudhary A, Kabra A, Rao MM, et al. Phytochemical and pharmacological properties of Withania somnifera: an updated review. Phytother Res. 2021;35(2):682–704.
17. El Omari N, Bakha M, Imtara H, Guaouguaou FE, Lyoussi B. Essential oils and neurodegenerative disorders: mechanisms of action and therapeutic prospects. Molecules. 2023;28(14):5418.
18. Abdulghani M, et al. Treatments of psychosomatic symptoms in Alzheimer's disease: a scoping review of the potential therapeutic effects of essential oils. Psychogeriatrics. 2025. doi:10.1111/psyg.70007.
19. Efficacy of Aromatherapy Against Behavioral and Psychological Disturbances in People With Dementia: A Meta-Analysis of Randomized Controlled Trials. J Am Med Dir Assoc. 2024;25(11):105199. doi:10.1016/j.jamda.2024.105199.
20. Hedigan F, Sheridan H, Sasse A. Benefit of inhalation aromatherapy as a complementary treatment for stress and anxiety in a clinical setting: a systematic review. Complement Ther Clin Pract. 2023;52:101750. doi:10.1016/j.ctcp.2023.101750.
21. Yoo O, Park SA. Anxiety-reducing effects of lavender essential oil inhalation: a systematic review. Healthcare (Basel). 2023;11(22):2978. doi:10.3390/healthcare11222978.
22. D'Andrea F, Tischler V, Dening T, Churchill A. Olfactory stimulation for people with dementia: a rapid review. Dementia (London). 2022;21(5):1800-1824. doi:10.1177/14713012221082377.
23. Banerjee A, Satish S, Pandya PH, Ramakrishna Shabaraya A. Aromatherapy in the treatment of Alzheimer's disease: a systematic review. Int J Pharm Sci Rev Res. 2021;66(2):93-97. doi:10.47583/ijpsrr.2021.v66i02.016.