

Aromatherapy: Ancient Solutions for Recent Problems

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ABSTRACT

Background: Currently, complementary and alternative medicine emerge as strategy in the prevention and treatment of various health problems. The present study aims to evaluate the efficacy of aromatherapy with *Lavandula angustifolia* Mill and *Matricaria recutita* L. in reducing stress and anxiety levels.

Methods: A study was performed with 36 individuals divided in two distinct groups: "aroma group" and "control group". Individuals of "aroma group" were treated with a mixture of essential oils of *Lavandula angustifolia* Mill and *Matricaria recutita* L. applied with *Effleurage* massage while the individuals of "control group" received only *Effleurage* massage. Stress and anxiety levels of all individuals participating in the study were evaluated before and after the treatment. Additionally, the chemical composition of the essential oils was evaluated by GC and GC-MS.

Results: In the aromatherapy assay, the "aroma group" showed a significant decrease of 12% and 30%, in stress and anxiety levels, respectively while lower decrease was observed in "control group" (3.3% and 2.6% for stress and anxiety levels, respectively).

Conclusion: In this study, aromatherapy was effective in stress and anxiety levels reduction and linalool, the major compound of the essential oil mixture, may be positively contributing for these effects.

Keywords: *Aromatherapy; Lavandula angustifolia* Mill; *Matricaria recutita* L.; *volatile oils; stress disorders; anxiety.*

1. INTRODUCTION

Natural products and particularly those from plants, are used by humans since ancient times with the purpose to relief and cure diseases or to maintain health [1]. Gradually the herbs and their essential oils became part of the prevention and treatment of diseases, particularly in situations where physicians frequently were not present, as in the case of rural areas. The use of medicinal plants for the treatment of various diseases was a common practice among the populations that resort to spontaneous plants due to easy local access [2,3]. The World Health Organization (WHO) recognizes several benefits in the use of complementary and alternative medicines (CAM) playing an important role in the treatment or prevention of chronic diseases and improving life quality [4,5]. One form of CAM, aromatherapy, comprises the use of essential oils as treatment, generally applied by inhalation, using a diffuser, aroma stones, adhesive patch or cotton balls [6,7,8,9], on the skin [10] or, less frequently, orally [11]. Today, aromatherapy is a form of treatment recognized by the WHO, commonly used to relieve pain, improve mood, and promote a sense of relaxation and used, between others, for sleep issues, and in anxiety daily stress problems [8].

The essential oils used in aromatherapy can have different origins, as they can be extracted from spontaneous plants or from cultivars, and their composition can vary with environmental factors, such as climatic and soil conditions [12,13,14]. Thus, it is very important to know the composition of the

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essential oils to compare and understand the relevance of the major compounds in the effects of aromatherapy [15].

The essential oil of *Lavandula* ssp. is well known and has been described as having antibacterial, antifungal, sedative, carminative properties and antidepressant activity [16]. Was also used in studies involving control of insomnia in midlife women [17], in students [18] and in cancer patient [19], which justify its use in aromatherapy and as pharmacological agent [20]. Studies about the benefits of *Lavandula* essential oils showed that linalool and linalyl acetate of this plant can stimulate parasympathetic system [21]. In addition, linalool and α -pinene are constituents of some essential oils with considerable antianxiety effects such as *Lavandula angustifolia*, *Piper guineense*, and *Cymbopogon citratus*, all of them have very clear anxiolytic-like effect [15].

Since a very long time Chamomile oil has been used as natural medicine and in aromatherapy [22]. Literature indicates *Matricaria recutita* L. essential oil as antispasmodic and anxiolytic agent [23] with moderate antioxidant and antimicrobial activities [24,25]. Inhalation of the chamomile flowers essential oils is recommended to relieve anxiety and depression.

In the present study, the participants' perceptions of aromatherapy were recorded. Additionally, we evaluate the effectiveness of essential oils used in aromatherapy in the reduction of stress and anxiety levels, by applying a mixture of essential oils (*Matricaria recutita* L. and *Lavandula angustifolia*) through *Effleurage* massage technique. The chemical composition of the essential oils was also determined.

2. MATERIALS AND METHODS

2.1 Collection of Plants and Essential Oil Extraction

Plants of *Matricaria recutita* L. were collected in the wild at Montesinho Natural Park, in the northeast of Portugal, during the flowering stage (July-August). Plants of *Lavandula angustifolia* Mill were collected in the gardens and greenhouses of Agrarian School of Bragança, between June and July. The species were identified, and registration vouchers deposited in Herbarium of the Escola Superior Agrária of Bragança - BRESA. The extraction of essential oils by hydrodistillation was performed using a Clevenger apparatus, for 3 h, according to the European Pharmacopoeia [26], with yield determination (v/fresh weight). The essential oils were stored at -20°C in the dark. From each essential oil, a small amount was used for the GC and GC-MS analyses and the remainder was used to prepare the mixtures for further use in the aromatherapy assays.

2.2 Aromatherapy Assay

The data were collected using a questionnaire, composed by three sections. In section 1, generic questions about the use of aromatherapy and sociodemographic questions were included. Section 2 was the perceived stress Cohen Perceived Stress Symptoms scale (1983) with 14 items [27]. Section 3 was composed of Hamilton Anxiety Rating scale (1959) [28] for measuring the anxiety levels.

Section 2 and section 3 were applied to the participants before and after the aromatherapy or *Effleurage* massage treatments while section 1 was applied just before the treatments. For the experimental part of the study, an opportunistic sample was used. Adults aged between 18 and 45 years, users of a clinic in Bragança, Portugal, with high levels of stress and with medium or severe anxiety were included in the study. In this way, the inclusion criteria were a stress level equal or greater than 75% and a minimum of 20 points on the anxiety scale [27,28]. Individuals with psychiatric conditions, pregnant women and individuals under any type of treatment for stress and anxiety were excluded from the study.

36 individuals of both sexes, aged between 18 and 45 years were included in the study. The participants, mostly of female gender (83%, n=30) with ages between 18 and 25 (67%, n=24), were divided into two distinct groups: "aroma group" (n= 18) and "control group" (n=18) (Fig. 1). "Aroma group" received a treatment with a mixture of the two essential oils (*Lavandula angustifolia* Mill and *Matricaria recutita* L.; 60:40 v/v) using sweet almond oil as vector and *Effleurage* massage, while for "control group" only *Effleurage* massage with sweet almond oil, an odorless oil, was applied. The

complete treatment of the two groups consisted of fourteen sessions (once a week) of thirty minutes each. The levels of stress and anxiety of the participants were assessed before and after treatment, in the two groups [27,28].

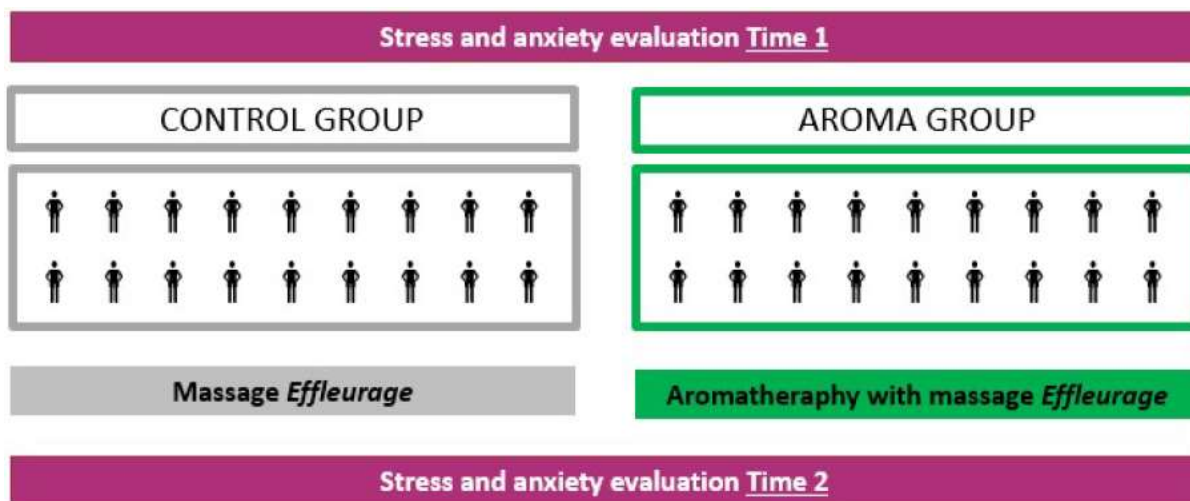


Fig. 1. Study design

During the study, all ethical aspects were guaranteed. Before starting, all participants were informed about the aim of the study, the procedures and the voluntary nature of their participation. They were informed that a refusal or a termination of their participation would not have any negative consequence, being able to withdraw from the study at any time. All participants have signed the informed consent form and, to ensure confidentiality, a code for each participant was used instead of their names, and the collective disclosure of results was also guaranteed.

2.3 Chemical Analysis of Essential Oils

Gas chromatography (GC): Gas chromatographic analyses were performed using a Perkin Elmer Autosystem XL (Perkin Elmer, Shelton, Connecticut, USA) gas chromatograph equipped with two flame ionization detectors (FIDs), a data handling system and a vaporizing injector port into which two columns of different polarities were installed: a DB-1 fused-silica column (30 m x 0.25 mm i. d., film thickness 0.25 μm) (J & W Scientific Inc., Rancho Cordova, CA, USA) and a DB-17HT fused-silica column (30m x 0.25mm i. d., film thickness 0.15 μm) (J & W Scientific Inc.). Oven temperature was programmed, 45-175°C, at 3°C/min, subsequently at 15°C/min up to 300°C, and then held isothermal for 10 min; injector and detector temperatures, 280°C and 300°C, respectively; carrier gas, hydrogen, adjusted to a linear velocity of 30 cm/s. The samples were injected using split sampling technique, ratio 1:50. The volume of injection was 0.2 μl of a pentane-oil solution. The percentage composition of the oils was computed by the normalization method from the GC peak areas, calculated as mean values of two injections from each essential oil, without using correction factors.

Gas chromatography-mass spectrometry (GC-MS): The GC-MS unit consisted of a Perkin. Elmer Autosystem XL (Perkin Elmer, Shelton, Connecticut, USA) gas chromatograph, equipped with DB-1 fused-silica column (30 m x 0.25 mm i.d., film thickness 0.25 μm) (J & W Scientific, Inc.), and interfaced with a Perkin-Elmer Turbomass mass spectrometer (software version 4.1, Perkin Elmer, Shelton, Connecticut, USA). Injector and oven temperatures were as above; transfer line temperature, 280°C; ion trap temperature, 220°C; carrier gas, helium, adjusted to a linear velocity of 30 cm/s; split ratio, 1:40; ionization energy, 70 eV; ionization current, 60 μA ; scan range, 40-300 amu; scan time, 1 s. The identity of the components was assigned by comparison of their retention indices, relative to C9-C17 n-alkane indices and GC-MS spectra from a homemade library, constructed based on the analyses of reference oils, laboratory-synthesized components and commercial available standards.

2.4 Statistical Analysis

The results were analyzed using the SPSS program (version 20) and the descriptive statistics expressed as mean, standard deviation and percentage. For statistic comparison the Wilcoxon and Mann-Whitney U tests have been used with a level of significance of 5% (p -value<0.05).

3. RESULTS AND DISCUSSION

3.1 Perceptions about Aromatherapy

Concerning aromatherapy treatment, the majority of participants were satisfied (67%) and 22% were totally satisfied (Fig. 2). In agreement with this, when questioned about the benefits of aromatherapy, most participants agreed that aromatherapy has health benefits, namely, it is useful (83%), promotes health (86%), is effective in preventing disease (86%), promotes well-being and quality of life (69%) and promotes a good relationship between the therapist and the client (83%).

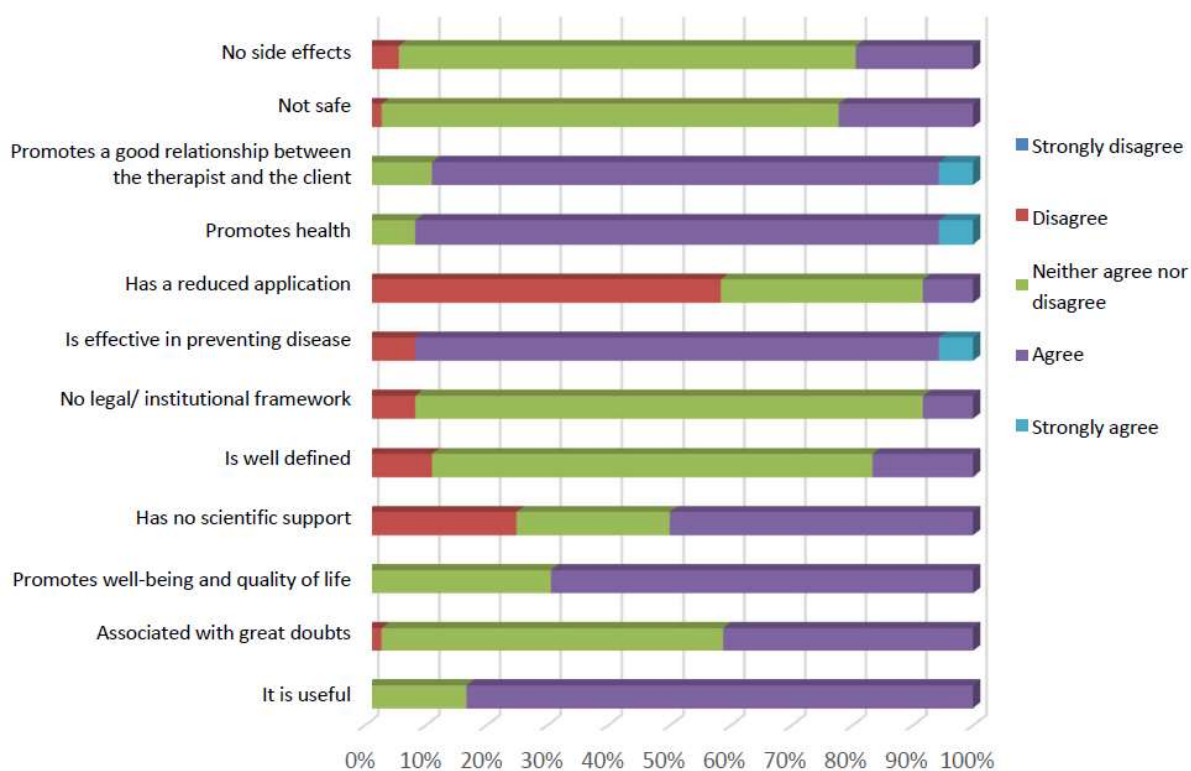


Fig. 2. Participants' perceptions of aromatherapy

Accordingly, 70% of the participants agreed with the inclusion of aromatherapy in the National Health Service, due to the following reasons (with percentages of response between 94 to 100%): lack of confidence in conventional medicine, less adverse effects by using natural products, efficient prevention of disease and the low cost of treatments.

A comprehensive view of the actual position of these participants in relation to aromatherapy has been collected, which recommended the inclusion of this therapy in National Health Service. In fact, the European Information Centre for Complementary & Alternative Medicine highlight the remarkable growth of these practices in the last ten years, meeting the recommendation that patients may have more choice outside the scope of conventional medicine [29, 30, 31].

3.2 Effects in Stress and Anxiety Levels

Regarding the levels of stress and anxiety in each group, there are statistically significant differences between the initial and final values of stress and anxiety of the two groups (Table 1). In the "aroma group" it was verified that the values distribution of "stress initial" and "stress final" are not identical. Similar results have been found for anxiety, with values of "anxiety initial" and "anxiety final" not identical.

Table 1. Initial and final levels of stress and anxiety

Variable	Aroma Group (n=18)				Control Group (n=18)				Aroma Group (n=18)	Control Group (n=18)	p-value ⁽²⁾
	Initial Level	Final Level	Test Values	p-value ⁽¹⁾	Initial Level	Final Level	Test Values	p-value ⁽¹⁾			
Stress (%)	77.4±3.5	68.3±7.2	-3.731	<0.001	72.7±2.8	70.3±2.7	-3.520	<0.001	9.2±6.9	2.4±1.6	< 0.001
Anxiety (points)	25.3±7.6	17.7±7.9	-3.290	<0.001	17.2±2.0	16.7±2.0	-2.530	=0.011	7.7±8.1	0.4±0.6	< 0.001

The levels of stress e anxiety initial and final are expressed as average ± standard deviation; Values of the test and p-value of Wilcoxon test⁽¹⁾; Differences of stress e anxiety levels in each group are expressed as average ± standard deviation; p-value of Mann-Whitney U test⁽²⁾.

For the "control group" the distributions of the values of the variables "stress initial" and "stress final" are not identical and similar results have been found for "anxiety initial" and "anxiety final" values (Table 1). In detail, for the "aroma group" it was evident a decrease in the stress levels (12%) and anxiety (30%) after the treatment, from a rating "high" to "medium", compared to the "control group" while the levels of stress and anxiety have decreased of 3.3% and 2.6%, respectively. In this way, in the "aroma group" it was observed that anxiety levels decreased more intensively than stress levels.

Further, with regard to differences between the initial and final values of stress and anxiety levels, after individual and group analysis, a significant decrease of 6.7% and 7.2 points it was observed for stress and anxiety levels, respectively (Table 1). Moreover, it is important to note that the differences between initial and final values were greater in the "aroma group".

The results indicated that aromatherapy is an effective treatment in the reduction of stress and anxiety levels. Specifically, the "aroma group" showed a significant decrease of 12% and 30%, with respect to stress and anxiety compared to the control group in which a minor decline in these levels (3% in stress and 2.6% in anxiety) was observed. The differences in stress and anxiety levels between aroma and control groups before and after treatment were also significant.

Similar studies have demonstrated the benefit of aromatherapy in stress control in primary school teachers [32] and nurses [33]. Additionally, various studies have shown that aromatherapy reduces anxiety in surgical patients [34, 35].

To clarify the aromatherapy regarding the effectiveness, it is important the increase of research in this area [36]. In addition, the monitoring and reporting of possible side effects should be done to ensure the safety of this technique [37, 38, 43, 44].

3.3 Chemical Composition of *Lavandula angustifolia* and *Matricaria recutita* Essential Oils

The essential oils of *Lavandula angustifolia* and *Matricaria recutita*, were obtained with a yield of 3.5% and 0.41% (v/fresh weight), respectively. The composition of the essential oils of Lavender, of Chamomile and of the mixture used in the aromatherapy assay is shown in Table 2, considering only the major compounds ($\geq 2\%$).

Table 2. Major compounds in the essential oils of Lavender, Chamomile and their mixture

Components	RI	Lavender	Chamomile	Mixture
α -Pinene	930	0.6	3.8	1.9
Isobutyl isobutyrate	999	-	2.6	0.3
1,8-Cineole	1005	7.5	13.9	12.2
Limonene	1009	1.3	-	2.0
<i>cis</i> - β -Ocimene	1017	5.0	-	4.1
Linalool	1074	45.1	0.5	43.4
Camphor	1102	8.6	0.3	8.2
<i>trans</i> -Pinocarveol	1106	-	4.7	-
Pinocarvone	1121	-	7.8	1.3
Borneol	1134	5.5	-	4.8
Terpinen-4-ol	1148	7.0	0.6	6.3
Linalyl acetate	1245	2.9	-	2.3
Hexyl angelate	1273	-	3.9	-

*RI: Retention Index relative to C₉-C₁₇ n-alkanes on the DB-1 column

For Lavender essential oil, with a percentage of identification near 99%, the more abundant compounds were linalool, camphor, 1,8-cineole, terpinen-4-ol, borneol and *cis*- β -ocimene. Although with a lower percentage of identification (approximately 50%) for Chamomile, the main compounds were 1,8-cineole, pinocarvone, *trans*-pinocarveol, hexyl angelate, α -pinene and isobutyl isobutyrate. In the case of Chamomile monoterpenes 1,8-cineole and α -pinene, presents in our analyses, it was register in the bibliography that they were able to decrease apoptosis by reduced capase-3 activity. These antioxidant action involve ROS scavenging and also induction of the nuclear Nrf2 factor. Was already demonstrates the potential beneficial therapeutic effect of these monoterpenes, on the oxidant/antioxidant balance, in different diseases of the central nervous system [39]. It is noteworthy the different profile of the two essential oils with some compounds present in the essential oil of only one of the plant species while others are common to both plant species essential oils, but frequently with clearly quantitative differences (e.g. linalool and terpinen-4-ol).

The essential oils analyses allow to found compounds such as linalool, 1,8-cineole in *Lavandula*. As described in the literature, the essential oil of this plant is mostly composed by oxygen-containing monoterpenes compounds [15]. Although the composition of the essential oil of *Lavandula angustifolia* shows a remarkable variability, depending on the source, the season or the method of isolation [40-44], the main constituents are predominantly linalool, 1,8-cineole or linalyl acetate, and oxygenated monoterpenes constitute, largely, the major group. A recent publication showed that the essential oil of Lavender and its main constituent, linalool, have a potential inhibitory effect on neuronal spontaneous electrical activity, using rat neuronal networks [45]. Linalool and linalyl acetate, produce an anxiolytic effect, via inhibition of voltage-gated calcium channels, reduction of 5-HT_{1A} receptor activity, and increased parasympathetic tone. In these studies, linalool may be the main active component of

lavender linalyl acetate, which showed no anxiolytic-like action by itself, but increased the anxiolytic effect of linalool [46].

4. CONCLUSION

In the present study, the participants have a positive opinion about aromatherapy. The study showed the importance of aromatherapy in the reduction of stress and anxiety levels, however more research is needed to understand which compounds are involved and the mechanism of action related to each essential oil used.

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CONFLICT OF INTEREST

The authors confirm that this article content has no conflicts of interest.

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