

Evaluate the Effect of Harvesting Age and Drying Methods on Essential Oil Yield of *Rosmarinus officinalis* L. Leaves

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ABSTRACT

Rosemary (*Rosmarinus officinalis* L.) is an attractive, evergreen, highly branched aromatic medicinal plant. It is used to roast meats, fish, potato salads to prevent food poisoning, spice for the preparation of food ingredients for pea mash, and pepper powders, traditional medicines and cosmetics in Ethiopia. Information about proper harvesting age and drying methods of this international valuable plant is essential. The objective of this study was evaluating the effect harvesting age and drying method on the essential oil content of rosemary leaves. The experiment consisted of three levels of harvesting ages (6, 12, and 18 MAT and four levels of drying methods (fresh, sun, shade, and oven) in a randomized complete design with three replications. The data were statistically analyzed using analysis of variance and values of least significant difference at 5% using SAS. The highest essential oil contents (0.959%, 2.289%, 0.84%, 2.01%; 0.8392%, 2.138%, 0.741%, 1.88%); while the lowest (0.5517%, 1.34%, 0.5017%, 1.2167%) obtained from 6, 12 and 18 MAT respectively. Therefore, harvesting age had a significant effect on the percentage change (53.94%, 52.3%, 50.42%, 49.05%; 41.34%, 45.86%, 38.48%, 43.05%) essential oil content increments were observed from 12 and 18 MAT when compared to 6 MAT, respectively. Drying methods has significant effects on essential oil contents (0.899%, 2.25%, 0.807%, 2.02%; 0.848%, 2.09%, 0.74%, 1.82%), while the lowest (0.689%, 1.702%, 0.6144%, 1.46%, 0.698%, 1.646%, 0.6156%, 1.504%) was obtained from fresh, shade, sun and dried leaves respectively. Therefore, drying methods had significant effects on the percentage change in essential oil content during the drying when compared with fresh (26.45%, 27.72%, 27.119%, 31.94%; 25.19%, 30.22%, 26.87%, 29.31%, and 5.85%, 7.32%, 8.62%, and 10.41%) from oven, sun and shade-dried leaves respectively. The interaction of harvesting age and drying methods had a highly significant ($p < 0.05$) influence on the essential oil content.

Keywords: Essential oil content; Harvesting age; Drying methods; *Rosmarinus officinalis*; Food poisoning

INTRODUCTION

Rosemary (*Rosmarinus officinalis* L.) is an aromatic medicinal plant that is attractive, evergreen, and highly branched. It can reach a height of one to two meters, has opposite branches, and thick, leathery leaves that are smooth, woolly, whitish, and glandular on the underside, with small clusters of flowers at the tips of the branches. It is the most important and popular crop planted in every Ethiopian home garden, and the majority of Ethiopians now plant in vast agricultural lands in agricultural extension systems in Silte, Garage, Wondo Genet, Wollo,

around Addis Abeba and other parts of Ethiopia. FBC also stated that rosemary has a number of applications for improving livelihoods in Ethiopia, including fresh leaves for roasting meats, fish, and potato salads to prevent food poisoning, spice for the preparation of food ingredients for pea mash, and pepper powders, traditional medicines, and cosmetics as cash crops [1].

Essential Oils (EOs) of rosemary leaves are economical products that are used in the fragrance, cosmetics, food, pharmaceutical, flavoring, soap, detergent, household sprays, shampoo, toilet soap, and pharmaceutical industries. The majority of Ethiopian

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women believe the assumption that rosemary essential oil can promote skin and hair follicle growth [2].

It is also well known for its antimicrobial properties, carminative properties, and ability to flavor meat, sauces, and condiments. It also has anti-inflammatory, astringent, carminative expectorant, nervine, emmenagogue, therapeutic, tonic, stimulant properties, improves blood circulation, helps ease headaches, and antibacterial and antifungal properties that aid in digestion and increase food absorption. They also stated that rosemary leaf essential oil has a strong effect on the brain because it clears the mind and aids concentration, and because it is an exceptional fixative material that contributes a strong, fresh odor that blends well with various other essential oils' odors and serves to mask the unpleasant smells of certain other ingredients [3].

Rosemary is a perennial crop that can live for five years or more, depending on the crops grown. The plant prefers light, dry soil with a pH range of neutral to alkaline, preferably in areas with rocky soil. The essential oil yield from this important plant is determined by the metabolic state of the leaf tissue responsible for secondary metabolism synthesis/natural products, which is closely related to the physiology of the plant's entire leaf maturity. The yield of essential oils in dried medicinal aromatic plant leaves is determined by the integrity of the oil glands and drying parameters such as the drying method, temperature, drying time, amount of water evaporated during drying, and distillation time of essential oil extractions [4].

Drying methods lead to the loss of essential oil because it is easily converted into other types of compounds when exposed to higher temperatures *via* chemical reactions such as oxidation, isomerization, cyclization, or dehydrogenation. A large number of herb-drying studies have been conducted in recent decades and several herb drying methods/conditions have been introduced. Mostly due to its bulky weight medicinal aromatic herbs and plants are sold in dried forms for spices at early age. However, there is no common understanding the effects of drying herbs and age of harvesting on essential yield of *Rosmarinus officinalis* leaves in Ethiopia. The same way, the dynamic interactions between primary and secondary metabolism are critical when considering the biosynthesis and accumulation of plant natural products/essential oils for aromatic plants. Even though market demand is increasing many folds in Ethiopian international markets, there has been no scientific study of the effect of drying methods and harvesting age on the yield of essential oil from *Rosemary officinalis* leaves in the study area [5].

Therefore, determining the optimal plant harvesting age and appropriate drying methods for essential oil extractions from rosemary leaves is important. The different studies agreed that the essential oil yield from dried herbs at younger ages is still lower than that of fresh herbs.

Therefore, it is essential to make people aware of how much the essential oil content changes during the drying process, storage period, and crop growth cycle through the use of metabolite changes in natural products' primary and secondary metabolites. The goal of this study was to see how these two factors affected the percentage of essential oil yield of *Rosmarinus officinalis* L.

leaves. Hence, this article may be useful in developing new possibilities for farmers, agricultural extension agents, governments, non-governmental organizations, and other small businesses in Ethiopia to engage in full-scale production of rosemary essential oil [6].

Objectives:

- To investigate the effect of harvesting age and drying methods on essential oil yield of *Rosmarinus officinalis* L. leaves.
- To investigate the interaction effect of harvesting age and drying methods on essential oil yield *Rosmarinus officinalis* L. leaves.

MATERIALS AND METHODS

Description of the experimental site

This study was conducted at the Ethiopian institute of agricultural research, Wondo Genet agricultural research center, from December 2019 to December 2020 for one year. The geographical location of the study area ranges from 38° 37'13"-38° 38'20" East and 7° 5'23"-7° 5'52" North with an altitude of 1780 m a.s.l. Southeast of Shashemene, about 14 km, and 267 km South of Addis Ababa. The site receives a mean annual rainfall of 1128 mm, with minimum and maximum temperatures of 11.47°C and 26.51°C, respectively. The soil textural class of the experimental site is sandy loam with a pH of 7.2 (Figure 1).

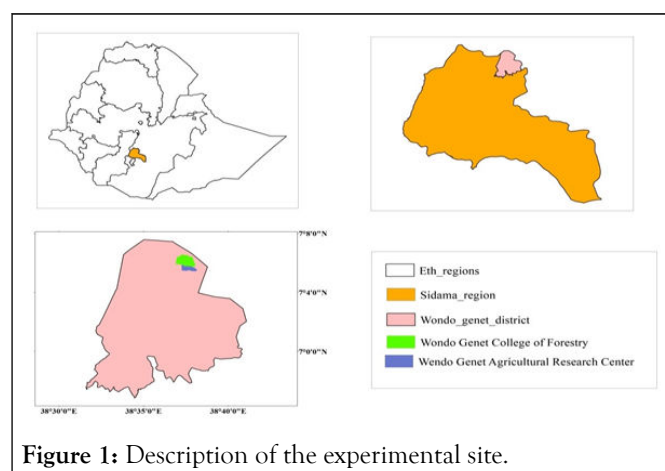


Figure 1: Description of the experimental site.

Plant data collection

The current study consisted of three levels of harvesting ages (6, 12, and 18 Months After Transplanting (MAT)) and four levels of drying methods (fresh, sun, shad, and oven drying) in a Randomized Complete Design (RCD) with three replications. Harvesting took place in the second half of April 2019, just as the plants from disease-free, uniformly grown seedlings of three age categories arrived at the Wondo Genet agricultural research center's station at the same time. Plant samples were collected by randomly selecting six plants from the central rows of each plot, excluding the borders (planting times were tagged on each plot) [7].

The collected plants were taken to the laboratory as soon as they were harvested, and the leaves were separated from the stems

and divided into four batches, one of which was fresh, and the other three batches were immediately dried using oven drying, sun drying, and shade drying methods. The plant data was collected in the form of the images below (Figure 2) [8].



Figure 2: Plant sample collection from d/t harvesting age (i.e. 6, 12 and 18 months after transplanting).

Drying procedure

Until the equilibrium moisture content was reached, the weight loss in all drying treatments was measured using an analytical balance. Because the three drying methods used different heat sources during the drying process, drying times varied accordingly: Shade>sun>oven. Overall, it took seven days of shade drying, five days of sun drying, and one day of oven drying to achieve the final moisture content (i.e., air dried at an average relative humidity of 23%-39%, sun dried at an average temperature of 25°C at the same relative humidity. Similarly, sun-drying rosemary leaves at 25°C for five days (herbs were exposed to direct sunlight) is sufficient, but it causes essential oil components to evaporate. For oven dried leaves, the leaf was dried at 40°C for 24 hours before being tested. The same literature confirmed that drying rosemary leaves for 24 hours at 40°C is preferable, but increasing the temperature from 50°C to 65°C causes an unpredictably large reduction of the essential oil content of rosemary leaves [9].

Shade/air drying was carried out in a well-ventilated room for 7 days (mean temperature, 25°C; mean relative humidity, 30%). A similarly, Tambunan, et al., study observed that air temperature was used to dry medicinal herbs for 5 days-7 days. Since oven drying maintains a constant temperature throughout the drying process, there was no temperature variation other than for a brief period when the electricity was turned off. The collected material was dried using the mentioned (fresh, sun, oven, and shade) (Figure 3).



Figure 3: The process of OD drying leaves (i.e. Fresh leaves extraction, sun drying, oven drying and shade drying as well as moisture content determinators).

Extraction of essential oils

The essential oil of rosemary leaves was extracted using a cleverger apparatus and hydro-distillation. A quantity of 300 g of fresh based leaves was dried, and the moisture content was calculated except for fresh-leaf oil extractions. The prepared sample was placed in a round bottomed flask containing 600 ml of distilled water for the extraction of essential oils from leaves by hydro-distillation under optimal operating conditions. The material was completely immersed in water and boiled for three hours on a heating mantle [10].

During the distillation process, vapour containing both essential oils and water vapour is passed to the condenser; the condenser converts this steam into liquid form and drops it into harvesting material with a lower density than water, which is then placed on top of the water. After three hours of overseeing the distillation, the droplet collection completely stopped, and two phases were observed: an aqueous phase (aromatic water) and a less dense organic phase (essential oil).

Therefore, the less dense material is easily removed by opening up the steam coil until almost all of the water is lost, then closing the steam coil with its closing mechanism. Following that, the essential oil was separated from the water by decanting it or skimming it off the top with a pressure-paced pipet, the most important piece of equipment used to take essential oil from top to bottom, and reading the amount and meticulously recording it on the datasheet. The essential oils were extracted as follows using a cleverger apparatus/hydrodistillation (Figure 4) [11].

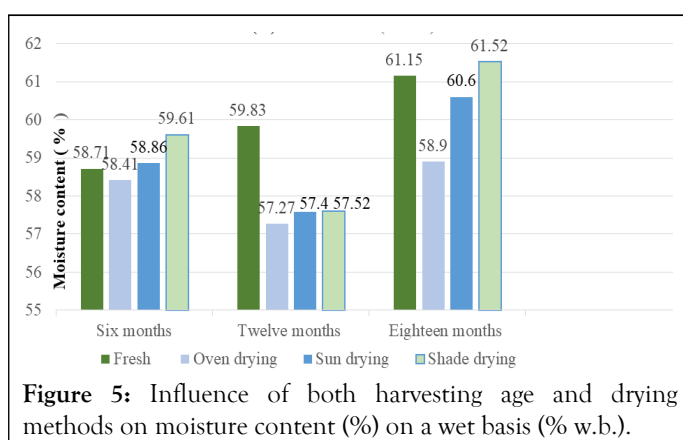


Figure 4: The process of extraction of essential oils by hydrodistillation using a cleverger apparatus.

RESULTS AND DISCUSSION

Moisture content

The Figure 5 below illustrates the moisture content of three harvesting age groups and three different drying methods. The percentage of moisture content of rosemary leaves was relatively highest (58.71%, 59.83% and 61.15%, and followed by 59.61%, 57.52% and 61.52% for fresh and shade-dried respectively, at six, twelve, & eighteen months after transplanting. Oven and sun dried leaves, on the other hand, had a lower (57.27%, 58.41%, 58.9%, 57.4%, 58.86%, and 60.6%) percentage of moisture content at six, twelve, and eighteen months after transplanting, respectively. Similarly, *M. citrata*, had higher moisture content in fresh leaves and air/shade dried leaves samples, but the percentage of moisture content was significantly affected by sun and oven drying [12].



Therefore, in this study, the essential oil content was significantly higher in fresh and shade-dried samples than in oven and sun dried samples, as shown in Tables 1 and 2 below. The comparison of the final moisture content demonstrated that the essential oil content was lost during the drying process. The essential oil content of dried leaves was statistically identical to the moisture content summarized in a graph. This finding is supported by Figiel, et al., investigation that water vapour can act as a carrier during the drying process, allowing volatile compounds to diffuse from tissues to the surrounding environment through evaporation. They also confirmed that moisture diffuses from the interior of the rosemary leaves to the surface during drying, bringing essential oil with it [13].

Therefore, it is clear that the moisture content is directly related to the essential oil content or volatile oil content loses during drying. This implies that additional to drying temperature decrease in volatile constituents and essential oils are associated with the amount of water removed during the drying process [14].

Table 1: The effect of harvesting age essential oil content of *Rosmarinus officinalis* L. leaves.

Harvesting age	EOC (%) V/W fresh based	EOC (%) V/W dry based	EOC (%) W/W fresh based	EOC (%) W/W dry based
Six MAT	0.5517	1.34	0.5017	1.2167
Twelve MAT	0.9592	2.2892	0.84	2.0075

The analysis of variance showed that harvesting age had a significant ($p < 0.05$) impact on essential oil content (Table 1). The highest essential oil contents (0.9592%, 2.2892%, 0.84%, and 2.0075%) were found rosemary leaves harvested from twelve months after transplanting, followed by 0.8392%, 2.1375%, 0.7408%, and 1.8842% for rosemary leaves harvested from and eighteen months after transplanting, volume by weight fresh based, volume by weight dry based, weight by weight fresh based, and weight by weight dry based, respectively. The lowest essential oil content (0.5517%, 1.34%, 0.5017%, and 1.2167%) was obtained from leaves harvested six months after transplanting volume by weight fresh-based, volume by weight dry based, weight by weight fresh based, and weight by weight dry based, respectively. Therefore, this research, harvesting age had a significant effect on the percentage change in essential oil content when compared to six months after transplanting (53.94%, 52.3%, 50.42%, 49.05%) essential oil content increments were observed twelve months after transplanting and (41.34%, 45.86%, 38.48%, 43.05%) essential oil content increments were observed eighteen months after transplanting volume by weight fresh based, volume by weight dry based, weight by weight fresh based, weight by weight dry based respectively. In particular, rosemary leaves harvested six months after transplanting yield less essential oil than those harvested twelve and eighteen months after transplanting. The same study reported that essential oils with a high water adhesion are more likely to evaporate during the drying process. Similarly, Zewdinesh, et al., found that the essential oil content of *Rosmarinus officinalis* L. leaves increases as harvesting age increases [15]. This is also in agreement with the results of Getachew and Aynalem, who observed that harvesting age had a highly significant effect on the essential oil yield of *Cymbopogon citratus*. Similarly, Solomon and Beemnet found that essential oil yield in spearmint and Japanese mint increases with harvesting age. They also stated that young rosemary leaves are small, immature, and thin, implying that high drying temperatures may have ruptured the oil glands, resulting in rapid evaporation of oil and essential oil losses during drying. Furthermore, six months after transplanting rosemary leaves, natural products were primarily used for growing primary metabolites. Hence, this study confirms that the harvest date was delayed, resulting in an increase in plant height, leaf length, leaf diameter, leaf thickness, leaf volume, and leaf number, and thus an increase in natural products or essential oil yield through a secondary metabolite accumulation of rosemary leaves on *Rosmarinus officinalis* leaves [16].

Eighteen MAT	0.8392	2.1375	0.7408	1.8842
LSD 0.05	0.0633	0.1369	0.0496	0.1235
CV	8.23	8.51	8.45	8.67

Note: MAT: Months After Transplanting; EOC: Essential Oil Content; V/W: Volume by Weight; W/W: Weight by Weight; CV: Coefficient Variations; LSD: Least Significant Difference.

According to Wondu, et al., the increased essential oil content of lemongrass with age might be due to plants evolving secondary metabolism to produce a diverse range of secondary metabolism and natural products *via* age. In contrast to this study, Tajidin, et al., and Maione found that the essential oil concentration of lemongrass (*Cymbopogon citratus*) dropped as the harvesting maturity stages. In our analysis, it was also proven that the production of essential oil began to decline a year after the plant was transplanted. Similarly, Tajidin, et al., observed that essential oil content in Lemongrass (*Cymbopogon citratus*) decreased when leaves harvested at too maturity stages. Mallavarapu, et al., reported a similar trained on *Artimisia pallens* in which essential oil content was higher at maturity of the leaves than at leaf initiation.

These secondary functions include essential functions for plant survival and development such as plant signaling and germination control, acting as a hormone for regeneration, and so on. Similarly, Amaducci observed a 16% increase in gel yield from early harvest to late harvest in chicory. Thus, the dynamic interactions between primary and secondary metabolism are critical when considering the biosynthesis and accumulation of plant natural products. As the result, the harvest date was delayed and the time duration for plant growth was increased, resulting in an increase in plant height, leaf length, leaf

diameter, leaf thickness, leaf volume, and leaf number, and thus an increase in natural products or essential oil yield. Similar, Yeşil and Zcan, who reported that as a plant grows more organic molecules, are stored beneath the trichomes on its leaves than are used as primary metabolites. Therefore, essential oil content increased with harvesting age, which could be attributed to the use of available resources such as light, nutrients, and water for the extraction of essential oil components for secondary metabolites through photosynthesis [17].

The below Table 2 shows that the drying method had a significant (p 0.05) effect on the essential oil content. The highest essential oil contents (0.8989%, 2.25%, 0.8067%, 2.0211%) were obtained from fresh leaves followed by (0.8478%, 2.0911%, 0.74%, and 1.8211%) obtained from shade dried rosemary leaves, while the lowest essential oil contents (0.6889%, 1.7022%, 0.6144%, 1.4644% and 0.6978%, 1.6456%, 0.6156%, 1.5044%) from sun and oven dried leaves (volume by weight fresh based, volume by weight dry based, weight by weight fresh based) respectively. This finding is consistent with findings that drying can result in a significant reduction in the amount of essential oil in many types of herbs, including basil (36%-45%), marjoram (23%-33%), and oregano (6%-17%), even when herbs are air dried at room temperature.

Table 2: The effect of drying methods on essential oil of *Rosmarinus officinalis* L. leaves.

Drying methods	EOC (%) V/W fresh based	EOC (%) V/W dry based	EOC(%) W/W fresh based	EOC (%) W/W dry based
Treatment				
Fresh (control)	0.8989	2.25	0.8067	2.0211
Oven drying	0.6889	1.7022	0.6144	1.4644
Sun drying	0.6978	1.6456	0.6156	1.5044
Shade drying	0.8478	2.0911	0.74	1.8211
LSD 0.05	0.0496	0.0573	0.1235	0.1426
CV (%)	14.98	15.08	14.58	14.43

Note: EOC: Essential Oil Content; V/W: Volume by Weight; W/W: Weight by Weight; LSD: Least Significant Difference.

This is uniform with the use of both sun and oven dried leaves, which produce the least oil, but fresh and air/shade dried leaves are adequate for oil extraction from rosemary leaves. This finding is also in line with previous research on the vast majority of medicinal aromatic plants. It was also demonstrated that drying in the shade at room temperature resulted in fewer volatile compound losses than sun and oven drying at 45°C.

Therefore, our findings make it clear that drying methods had significant effects on the percentage change in essential oil

values that were lost during the drying of leaves when compared to fresh (control): 26.45%, 27.72%, 27.119%, and 31.94% from oven dried leaves; 25.19%, 30.22%, 26.87%, and 29.31% from sun dried leaves; and 5.85%, 7.32%, 8.62%, and 10.41% from shade dried rosemary leaves (volume by weight fresh based, volume by weight dry based, weight by weight fresh based and weight by weight dry based) respectively. However, no significant difference in essential oil content occurs between sun and oven-dried leaves across all parameters. Similarly, when basil was dried in an oven at 45°C and in the sun at 25°C, the volatile

components were reduced by 19% while the essential oil was lost in an equal amount. Nevertheless, the essential oil content of both fresh and shade-dried rosemary leaves is higher than that of both oven and sun dried leaves. This study confirmed Ibanez, et al., findings that rosemary essential oil content dried in air or shade was most similar to fresh rosemary leaves essential oil content [18].

It is supported by Choe and Min, who state that heat facilitates the formation of free radicals during the drying process of aromatic herbs, allowing the essential oil to evaporate easily. Similarly, Mensure, et al., states that drying herbs at high temperatures affects the quantity and quality of essential oil. They also believe that the temperature sensitivity of the oil constituents is due to structural changes caused by drying, and that the majority of the oil loss occurred at the beginning of the drying process.

This study demonstrated that drying changed the surface structure of the leaves, and in some cases, they ruptured during drying, releasing volatile compounds that are expected to evaporate, resulting in essential oil reductions with respect to the maturity of leaves harvested Barिताux. Similarly, Mohammad, et al.; Xing, et al.; Tambunan, et al.; Rohloff, et al., observed that increasing the air temperature to 50°C reduced the essential oil content of rosemary leaves. It also agreed with, who investigated that the essential oil content of *L. citriodora* decreased as drying temperatures increased. Other researchers found that most medicinal plants are sensitive to high temperatures during drying. They demonstrated that increasing the drying temperature of medicinal plants can harm their glandular trichomes, which contain natural products. Similarly, fresh and shade drying, on the other hand, are ideal for preserving limonene and essential oil extraction. The same study confirmed that drying leaves at 30°C and 45°C resulted in 16%

and 23% loss of essential oil, respectively, whereas drying at higher temperatures resulted in significant essential oil losses, such as 65% at 60°C. This is supported by the findings of Jaganmohan, et al., who investigated that volatiles in rosemary dried in a 45°C oven lost 17% of their volatiles while drying in a microwave oven lost 61% of their volatiles. Therefore, this study proved that raising the drying temperature causes a greater loss of essential oil content. So, for essential oil extractions from *Rosmarinus officinalis* leaves, fresh and shade-dried rosemary leaves are recommended [19].

The highest value of essential oil content (1.0567%, 2.72%, 0.93%, 2.3967% from fresh leaves harvested from 18 MAT, followed by 1.0233%, 2.5433%, 0.8967%, 1.0567% from fresh leaves harvested from 12 MAT; while the lowest value was obtained at the harvesting age of six months after transplanting (volume by weight fresh based, volume by volume dry based, weight by weight fresh based, weight by weight dry based) under all dried conditions (fresh, shade, oven, and sun dried leaves), respectively (Table 3). Therefore, the variance analysis showed that the interaction of harvesting age and drying methods had a highly significant ($p < 0.05$) influence on the essential oil content of *Rosmarinus officinalis* leaves. Furthermore, it has been illustrated that matured rosemary leaves contain more essential oil than younger leaves from all drying methods. It is supported by Ascrizzi, et al., findings that the changes in volatile components during the drying process are also dependent on biological factors of the herbs, such as initial moisture content, plant age, growth conditions, harvesting time, and distillation time.

Table 3: Mean comparison of interaction between harvesting age and drying methods on essential oil content of *Rosmarinus officinalis* L. leaves.

Treatment	6 Months After Transplanting (MAT)			
	EOC (%) V/W fresh based	EOC (%) V/W dry based	EOC (%) W/W fresh based	EOC (%) W/W dry based
Fresh based	0.6167	1.4867	0.5933	1.43
Oven drying	0.55	1.32	0.4967	1.19
Sun drying	0.5167	1.2533	0.4667	1.1533
Shade drying	0.5233	1.3	0.45	1.0933
LSD 0.05	0.1097	0.2738	0.0992	0.247
CV (%)	8.27	8.41	8.44	8.57
Treatment	12 Months After Transplanting (MAT)			
	EOC (%) V/W fresh based	EOC (%) V/W dry based	EOC (%) W/W fresh based	EOC (%) W/W dry based
Fresh based	1.0233	2.5433	0.8967	1.0567
Oven drying	0.9067	2.12	0.7933	0.61
Sun drying	0.7467	1.7533	0.6567	0.83
Shade drying	1.16	2.7400	1.0133	0.86
LSD 0.05	0.1097	0.2738	0.0992	0.1097
CV (%)	8.27	8.41	8.27	8.27
Standard Error	0.0529	0.132	0.0478	0.0529
Treatment	18 Months After Transplanting (MAT)			

	EOC (%) V/W fresh based	EOC (%) V/W dry based	EOC (%) W/W fresh based	EOC (%) W/W dry based
Fresh based	1.0567	2.72	0.93	2.3967
Oven drying	0.61	2.2333	0.5533	1.35
Sun drying	0.83	2.1	0.74	1.8667
Shade drying	0.86	2.4967	0.74	1.9233
LSD 0.05	0.1097	0.2738	0.0992	0.247
CV (%)	8.27	8.41	8.44	8.57

Note: MAT: Moths After Transplanting; EOC: Essential Oil Content; V/W: Volume by Weight; W/W: Weight by Weight; CV: Coefficient of variations; LSD: Least Significant Difference

Therefore, our findings make it clear that drying methods had significant effects on the percentage change in essential oil values that were lost during the drying of leaves when compared to fresh (control): 26.45%, 27.72%, 27.119%, and 31.94% from oven dried leaves; 25.19%, 30.22%, 26.87%, and 29.31% from sun dried leaves; and 5.85%, 7.32%, 8.62%, and 10.41% from shade dried rosemary leaves (volume by weight fresh based, volume by weight dry based, weight by weight fresh based and weight by weight dry based) respectively. In the same way, twelve months after transplanting (weight by weight) and eighteen months after transplanting essential oil content was also affected at volume by volume dry based, weight by weight fresh based, and volume by volume dry based except volume by weight fresh based (Table 3).

However, there is a small statistically significant difference in the essential oil content of rosemary leaves dried in the oven versus the sun with each harvesting age. Similarly, essential oil compounds may be lost during the drying process due to various chemical reactions. Even though the harvesting age was increased from 6 months to 12 months after transplanting, the value of the essential oil content recorded increased with all drying methods, but the essential oil content began to decline after twelve months of transplanting, indicating that the leaves' age has reached its maximum storage potential for the essential oil contents [20].

The mean comparison of the interaction of drying methods and harvesting ages demonstrated that rosemary leaves dried early stage had significantly lower essential oil content than rosemary leaves dried at older age (Table 3). Younger rosemary leaves have less mature leaves, are greenish yellow in color, have less moisture content, and have thin layers with a high water affinity indicating that high drying temperatures may have ruptured the oil glands, resulting in rapid evaporation of oil and essential oil losses during drying, and the plants used natural products/primary metabolites) for growing rather than secondary metabolites (essential oil storage). It is supported by Ascrizzi, et al, findings that the changes in volatile components during the drying process are also dependent on biological factors of the herbs, such as initial moisture content, plant age, growth conditions, harvesting time, and distillation time. The same study observed that essential oils with a high water affinity are more likely to be lost during the drying process. Similarly, secondary products may constitute a high percentage of the total volatile content, accounting for more than 50% of the total volatile content.

This research found that there is no significant increment in yield after twelve months, despite the fact that production has reduced after twelve months of transplantation. Therefore, it is necessary to conclude that harvesting rosemary leaves for essential oil extractions using fresh and shade/air drying methods is appropriate twelve months after transplanting. Similarly, Menşure, et al., states that drying herbs at high temperatures affects the quantity and quality of essential oil. They also believe that the temperature sensitivity of the oil constituents is due to structural changes caused by drying, and that the majority of the oil loss occurred at the beginning of the drying process. This study demonstrated that drying changed the surface structure of the leaves, and in some cases, they ruptured during drying, releasing volatile compounds that are expected to evaporate, resulting in essential oil reductions by structural changes occurring during hot air drying.

CONCLUSION

Therefore, we concluded that both harvesting age and drying method had a statistically significant ($p < 0.05$) effect on *Rosemarinus officinalis* leaves essential oil content. The percentage of moisture content of rosemary leaves was relatively highest (58.71%, 59.83% and 61.15%, and followed by 59.61%, 57.52% and 61.52% for fresh and shade dried respectively, at six, twelve, and eighteen months after transplanting. Oven and sun dried leaves, on the other hand, had a lower (57.27%, 58.41%, 58.9%, 57.4%, 58.86%, and 60.6%) percentage of moisture content at six, twelve, and eighteen months after transplanting, respectively. The highest essential oil contents (0.9592%, 2.2892%, 0.84%, and 2.0075%) were found rosemary leaves harvested from twelve months after transplanting, followed by 0.8392%, 2.1375%, 0.7408%, and 1.8842% for rosemary leaves harvested from and eighteen months after transplanting while, the lowest essential oil content (0.5517%, 1.34%, 0.5017%, and 1.2167%) was obtained from leaves harvested six months after transplanting respectively. Therefore, this research, harvesting age had a significant effect on the percentage change in essential oil content when compared to six months after transplanting (53.94%, 52.3%, 50.42%, 49.05%) essential oil content increments were observed twelve months after transplanting and (41.34%, 45.86%, 38.48%, 43.05%) essential oil content increments were observed eighteen months after transplanting respectively. This study also investigated the influence of drying method on the essential oil content of rosemary leaves.

RECOMMENDATION

The highest essential oil contents (0.8989%, 2.25%, 0.8067%, 2.0211%) were obtained from fresh leaves followed by (0.8478%, 2.0911%, 0.74%, and 1.8211%) obtained from shade dried rosemary leaves, while the lowest essential oil contents (0.6889%, 1.7022%, 0.6144%, 1.4644% and 0.6978%, 1.6456%, 0.6156%, 1.5044%) from sun and oven dried leaves (volume by weight fresh based, volume by weight dry based, weight by weight fresh based) respectively. Therefore, our findings make it clear that drying methods had significant effects on the percentage change in essential oil values that were lost during the drying of leaves when compared to fresh (control): 26.45%, 27.72%, 27.119%, and 31.94% from oven-dried leaves and 25.19%, 30.22%, 26.87%, and 29.31% from sun-dried leaves; and 5.85%, 7.32%, 8.62%, and 10.41% from shade dried rosemary leaves (volume by weight fresh based, volume by weight dry based, weight by weight fresh based and weight by weight dry based) respectively. The variance analysis (Table 3) showed that the interaction of harvesting age and drying methods had a highly significant ($p < 0.05$) influence on the essential oil content of *Rosemarinus officinalis* leaves. Therefore, this study found that raising the drying temperature and harvesting rosemary leaves earlier results in a greater loss of essential oil content. Fresh and shade dried rosemary leaves with optimum harvesting age (after twelve transplanting) are recommended for essential oil extractions rosemary leaves essential oil. So, essential oils are volatile and must be handled with care and attention. Gas Chromatography-Mass Spectrometry (GC-MS) analysis is required for the physicochemical characterization of essential oils as well as the identification of components that evaporate during the drying process and secondary metabolites that develop with the age of rosemary leaves. Therefore, further research is needed to determine the rate of evaporation of essential/volatile oils and ingredients that evaporate quickly when dried *Rosemarinus officinalis* leaves are heated at high temperatures and determine the primary and secondary metabolites that emerge during the crop's life cycle and correlate them to essential oil production.

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