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Impact of microwave vacuum drying on physicochemical characteristics of mint (*Mentha spicata.* L) Leaves

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Abstract
Microwave Vaccum Drying (MVD) seems to be the best option for drying heat-sensitive food products, mainly leafy vegetable products. Spearmint (*Mentha spicata*) is principally known for its functional and medicinal food properties. In this study, the physicochemical characteristics of mint leaves are explained by drying in MVD. This is an assisted drying method that scopes to improve the quality of the product by preserving micro-nutrients. Here, three different microwave power levels (450 W, 630 W, and 810 W) and a vacuum of about 650 mm/Hg were used to investigate the physical and chemical characteristics of spearmint leaves. Data were analysed using one-way ANOVA. Maximum TPC and DPPH radical scavenging activity were attained at 630 W microwave power.

Keywords: *Mentha spicata*, microwave vacuum drying, chlorophyll content, total phenolic content, antioxidant activity

1. Introduction

Mint leaves are aromatic herbal plants belonging to the genus *Mentha* and consist of different varieties such as pineapple mint (*M. suaveolens*), spearmint (*Mentha spicata*), ginger mint (*M. gracilis*), peppermint (*M. piperita*), water mint (*M. aquatica*), horsemint (*M. longifolia*), and wild mint (*M. arvensis*) (Venkatachalam *et al.*, 2020) [21]. Spearmint has many medicinal characteristics to prevent headache, cough, and abdominal pain; hence, these leaves and their stem and flowers were brewed as herbal tea. It is also used in many manufactured food products, cosmetic products, and Asian cuisine as spice blends because of its antioxidant and antimicrobial activity. The mint essential oil is rich in Carvone compound, which is responsible for antioxidant and antimicrobial activity (Mohammadhosseini *et al.*, 2021) [15]. The moisture content of spearmint leaves at the time of plucking was about 75 to 95% (wb) (Muthukumar & Venkatesh, 2014) [16]. Hence, these leaves are perishable, leading to essential oil oxidation by the lipoxidase enzyme (Mahendran *et al.*, 2021) [13]. During storage, the mint leaves produce an unpleasant aroma, wilting, and reduction in nutritional value. This can be prevented by reducing the moisture content by up to 11%, slowing down the enzymatic reactions.

Drying is the most important method to reduce the moisture content and to preserve the mint leaves from bacterial and mold growth (Kennedy, 2016; Lavanya *et al.*, 2019) [12, 14]. The selection of a suitable drying method concerning the product to be dried is an important step as the method and operating conditions affect the dried product quality (Ghosh & Venkatachalapathy, 2014a, 2014b) [6-7]. The drying potential of the system can be improved either by raising the temperature at a given absolute humidity, decreasing the absolute humidity at a given operating temperature, or combining both (Naik *et al.,* 2022; Vijayan *et al.*, 2017) [20, 22]. The fresh mint leaves have a very short life period, up to 2 to 3 days. Drying leads to moisture reduction and also increases the quality of the leaves to the fresh leaves. Commonly used drying methods for drying *Mentha spicata* leaves include sun-drying, hot air oven drying, and tray drying. This conventional drying method leads to some disadvantages, such as loss in quality by degradation of heat liable nutrients, increased drying time, and final product contamination (Kilic, 2017) [11]. To overcome this drawback, novel drying techniques or non-thermal-assisted drying methods have to be used.

In that case, microwave drying can be used for drying mint leaves which leads to maximum thermal conductivity and an energy-saving system than other conventional drying methods. In some cases, the rapid drying of microwave drying can cause structural damage and hot spot because of their non-uniform electric field distribution (Lavanya *et al.*, 2019; Rao *et al.*, 2021) [14, 24].

To overcome these disadvantages, microwave-assisted vacuum drying has been introduced to dry agricultural fruits and vegetables (Bui-Phuc *et al.*, 2020) [4]. In Microwave Vacuum drying, the microwave energy was combined with a vacuum system for fast, low-temperature drying with maximum retention of bioactive compounds (JP *et al.*, 2021; Puttalingappa *et al.*, 2022) [10, 8]. This drying technique has maximum mass transfer at low temperatures, increasing the pressure gradient between the inside and outside layers. In the present study, the novel drying method, the microwave-vacuum drying technique, was used for drying mint leaves in less drying time and increasing the quality of dried mint leaves. Dried mint leaves have great economic importance (Karakaplan *et al.*, 2019) [9]. Since the drying method decides the quality of the dried final product, this study aims to determine the effect of microwave vacuum drying of dried mint leaf powder on the physicochemical parameters, which determines the good quality achieved using this drying method.

2. Materials and Method
2.1 Raw Material
Fresh mint leaves (*Mentha spicata.* L) Were procured from the local market at Thanjavur, Tamil Nadu. Manually the debris and soil from the surface of the leaves were discarded and cleaned in distilled water. Then the excess surface moisture water was removed by blotting paper. All the chemicals used were analytical grade.

2.2 Drying process
For microwave vacuum drying (MVD), a lab-scale microwave oven (Model: IFB 30SC4) with frequency-2450MHz, an output power of 0.9 kW. It consists of a vacuum pump (Value double-stage vacuum pump (VE215 N)), a glass vacuum desiccator, a hose pipe, a pressure regulator, an air-drying unit (with conical flask with silica gel sachets), and a vacuum gauge. The mint leaves were placed inside the glass vacuum desiccator mounted through a hose pipe with the vacuum pump. The vacuum pressure inside the drying system was controlled with the pressure regulator. The mint samples were dried at different microwave power levels of about 450W, 630W, and 810 W, and the vacuum level of about 650 mm Hg was maintained in the drying system. The air-drying unit entraps the water vapor during drying and ensures that the vacuum level is maintained. MVD-dried mint leaves were packed in ziplock covers and stored in a desiccator till further analysis. The dried mint powder was analysed to estimate its physical characteristics, such as color and water activity and chemical characteristics, such as total phenolic content, chlorophyll content, vitamin c content, and antioxidant activity.

2.3 Physical characteristics of dried mint leaves
2.3.1 Color
The color of the dried mint leaves was estimated using the Hunter color lab flex meter (Hunter Association Laboratory, Inc., USA). The $L*$ value indicates the lightness index ranges from 0 (black hue) to 100 (white hue), the * value represents the red to green color, and the $b*$ value indicates from yellow color to blue color (Naik *et al.*, 2021) [17]. The total color difference (ΔE) of the dried samples was calculated using the formula (1).

$$\Delta E = \sqrt{(L* - L_1*)^2 + (a* - a_1*)^2 + (b* - b_1*)^2} \qquad (1)$$

Where,
$L*$, $a*$, $b*$ are the dried sample color values at time t; L_1*, a_1*, b_1* are the fresh sample color.

2.3.2 Water activity
The water activity (a_w) of the dried mint leaves was estimated using the Aqua Lab Water Activity Meter (4TE).

2.4 Chemical characteristics of dried mint leaves
2.4.1 Total phenolic content
The total phenolic content of the dried mint extracts was determined by a UV spectrophotometer (Shimadzu-UV1800, Japan) using an oxidizing agent Folin–Ciocalteau Reagent (FCR). About 300 µL of methanolic sample extract was taken in a test tube and mixed thoroughly with 10% FCR of 2250 µL. Then 6% sodium carbonate solution of about 2250 µL was added after 5 min incubation. Finally, the extract mixture was kept at room temperature for about 90 minutes, and the total phenolic content was measured at 725 nm. The total phenolic compound was expressed as Gallic acid equivalent (GAE) in mg/g of sample extracts (Chumroenphat *et al.*, 2021) [5].

2.4.2 Chlorophyll content
The chlorophyll content in the mint extract was determined according to the study (Pandey *et al.*, 2015) [18] with minor modifications. About 0.5g of mint powder mixed with 80% acetone of about 20 ml. Then the mixture was centrifuged for about 5 min at 5000 rpm, and the supernatant was collected. This procedure was repeated until the color less residue was obtained. Finally, the chlorophyll content was measured at 663 nm and 645 nm in a spectrophotometer (Shimadzu-UV 1800, Japan). The chlorophyll content was calculated using the absorption coefficients.

2.4.3 Vitamin C content
A volumetric method was used to determine the vitamin c content in the mint sample extracts (Haldhar *et al.*, 2015) [6]. The working standard of 100 µg/mL was prepared by taking 10mL stock solution and diluted with100mL 4% oxalic acid, which was taken about 5mL and added with the 4% oxalic acid of about 10mL. The dye solution was prepared with a small amount of distilled water in 42mg sodium carbonate and added with 2, 6-dichloro phenol indophenol of about52mg. Next, the mixture was titrated with the dye solution. The display of pale pink color for about a few minutes is the endpoint. In the same way, about 0.5-5g of the methanolic sample extract was taken, and 4% oxalic acid was added, which was made up to 100 mL and allowed for centrifugation. About 5mL supernatant was mixed with 4% oxalic acid of 10 mL and titrated with the dye solution. The vitamin c content was expressed as mg/100g sample and calculated by the formula (2).

$$Vitamin\ C\ (mg/100g\ sample) = [(\frac{0.5mg}{V_1}) \times (\frac{V_2}{5}) \times (\frac{100}{sample\ weight}) \times 100] \quad (2)$$

V_1 is the dye volume used for standard (mL), and V_2 is the dye volume used for sample (mL).

2.4.4 Antioxidant activity
The antioxidant activity of the methanolic extracts of the mint was measured based on the capability to scavenge 2,2_-diphenyl-1- picrylhydrazyl stable radical (DPPH). With minor modifications based on (Maizura *et al.*, 2011) [14], about 200 µL of the mint extract was added with 0.1mM methanolic DPPH solution of 20mL and methanol of about 800 µL. Finally, the sample mixture was incubated for about 1h. The absorbance of the sample was measured using a spectrophotometer (Shimadzu-UV 1800, Japan) at 517 nm for all the samples. The scavenging ability of the DPPH radical was determined using the formula (3).

$$DPPH\ activity = 100 - \frac{Absorbance\ of\ sample - Absorbance\ of\ blank}{Absorbance\ of\ control} \times 100 \quad (3)$$

2.5 Statistical analysis
Software SPSS statistics 20.0 (International Business Machines Corp., USA) for analysis of variance (multivariate ANOVA) was used to determine the significant difference between the result data obtained. All the experiments were done in triplicates. The statistically significant for all the values were obtained at a 95% confidence level (p<0.05). Data were updated as mean with their standard deviation (Mean ± std. deviation). Excel charts were used (MS Excel 2017, Microsoft) for graph plotting.

3. Results and Discussion
3.1 Effect of microwave vacuum drying on the color and water activity of the mint leaves

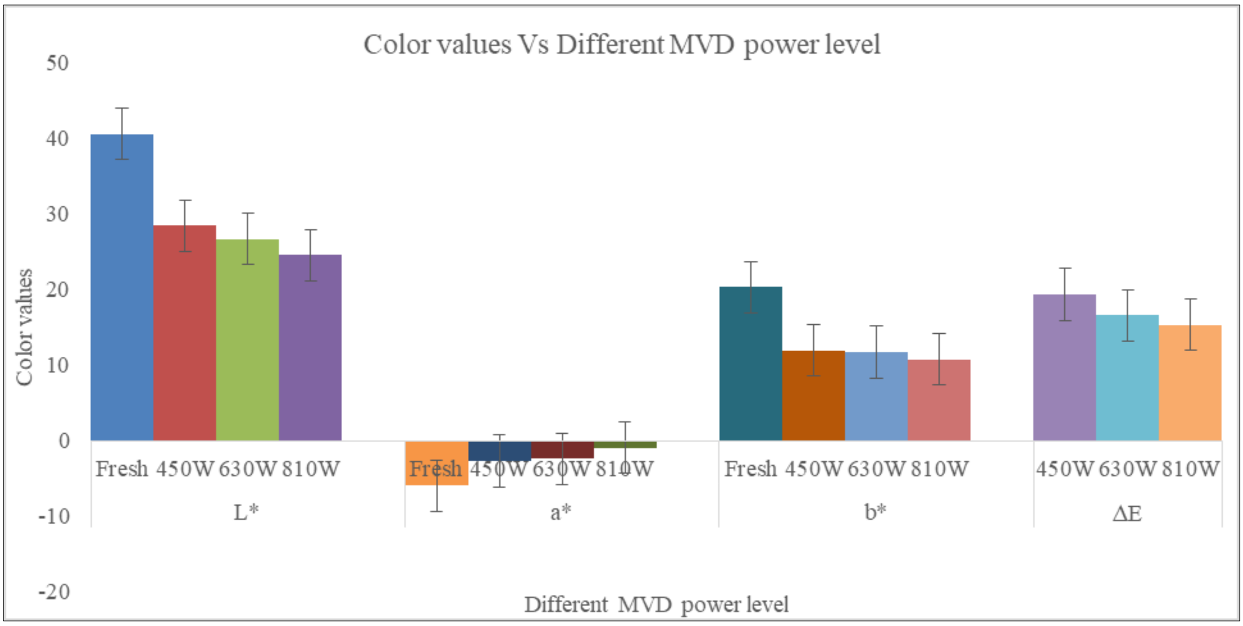

Fig 1: The graph plot between the color values and different microwave power levels.

The color characteristics obtained for the fresh and MVD-dried mint leaves are shown in Fig1. The fresh mint leaves were found to have an L^* value (brightness) of 40.55, a^* value (greenness) of -5.963, and b^* value (yellowness) of 20.28, respectively. Based on the results, for increasing the drying power level, the lightness of the mint leaves was reduced (the dark green leaves), and the lightness level was minimum at the highest power level, 810W. The change in color characteristics of the mint leaves depended on their different drying time, different power, and oxygen level. The chlorophyll in mint leaves was converted rapidly into pheophytins because of the increased drying time and high-power level (Ali *et al.*, 2014) [2]. During the drying process, the magnesium compounds responsible for the green color of mint leaves were replaced by hydrogen molecules (Beigi, 2019) [3]. The minimum conversion was found for the mint leaves dried at a low power level and less time. The results showed that the higher power level affects the greenness (chlorophyll) and the total color of the mint leaves.

The water activity (a_W) of the fresh mint leaves and MVD dried mint leave was 0.9718 to 0.4122. The results tabulated in Table 1 show that the water activity of different MVD-treated mint leaves has no significant difference. Since the water activity of the dried mint leaves was 0.4348, they are regarded as safe from microbial spoilage during storage.

3.2 Effect of microwave vacuum drying on chemical characteristics
3.2.1 Chlorophyll content of the mint leaves
The essential compound for photosynthesis is chlorophylls (Johan *et al.*, 2014) [7]. The chlorophyll content ranged from 91.110±0.716 mg/g to 87.645±0.813 mg/g. The mint leaves dried at 810W microwave power level showed high chlorophyll content (91.110±0.716 mg/g) followed by 90.574±0.370 mg/g dried at 630W, and 87.645±0.813 mg/g chlorophyll content for mint leaves dried at 450W. The color

value was proportional to the chlorophyll content in which the 450W dried mint leaves had light green leaves with low chlorophyll content, whereas a darker green color with the 810W dried sample had high chlorophyll content. The chlorophyll value of the mint leaves significantly varied based on the different power levels in MVD.

3.2.2 Vitamin C

Vitamin C is a heat liable compound that disintegrates at higher temperatures. The Vitamin C content of MVD dried at different power levels differs from 0.1806±0.003 to 0.4343±0.005 mg/100 g. Maximum Vitamin C was observed in the mint samples dried at 450W (0.4343±0.005 mg/100 g), followed by 630W (0.2136±0.416 mg/100 g) and finally 810W (0.1806±0.003 mg/100 g) power level. The fresh mint leaves have a vitamin C of about 31.8 mg/100g sample. It was found that the mint leaves dried at a low power level of 450 W showed high Vitamin C content. If no changes in Vitamin C content were found, then the other micronutrients present in the samples are also not affected by the drying process (Tanvir *et al.*, 2017) [17]. In this study, the high-power level 801W showed decreased Vitamin C content. From this result, it can be concluded that an increase in power level and a decrease in Vitamin C content were observed.

3.2.3 Total phenolic content and antioxidant activity of the Mint leaves

In herbs, the total phenolic content (TPC) is an important contributor to antioxidant activity. Hence TPC is responsible for the radical scavenging activity (Uribe *et al.*, 2016) [20]. DPPH radical scavenging activity and TPC content in this work were 20.034±0.467[a]% and 28.956±0.91[a] mg/g dried leaves**.** From the gallic acid calibration curves, the TPC was estimated. Antioxidant activity was expressed as radical scavenging activity (Al-Juhaimi[1] & Ghafoor, 2011; Lupsor *et al.*, 2019) [1, 12]. The result showed a significant increase in antioxidant activity and TPC for every MVD dried mint sample. The maximum antioxidant activity and TPC were found in the mint samples dried at a 630W power level. The antioxidant activity and TPC was 20.034±0.467[a] (%) and 28.956±0.91[a] (mg/g dried leaves) for the mint leaves dried at 630W, respectively. Under the drying process, the phytochemical bound inside the cell matrix is released, leading to higher antioxidant activity and TPC with a significant difference. Similarly, the maximum antioxidant activity and TPC were observed to be maximum for the mint leaves dried at 650W MVD.

Table 1: Water activity, TPC, and antioxidant activity of different microwave power level dried mint samples

Different microwave power	Water activity	Total phenolic content (mg/g dried leaves)	Antioxidant activity (%)
450W	0.4122±0.006[c]	18.100±0.875[c]	14.981±0.276[c]
630W	0.4173±0.002[b]	28.956±0.91[a]	20.034±0.467[a]
810W	0.4758±0.003[a]	24.938±0.596[b]	17.284±0.355[b]

Note: The means in the column at different superscripts were significantly different at $p<0.05$. (a–c) represents the significant ($p<0.05$) effect of drying power level on the physicochemical characteristics of mint leaves

4. Conclusion

This study analysed the physical and chemical characteristics of the mint leaves dried in a lab-scale microwave vacuum dryer at three different power levels. The minimum change in total color was observed in the mint leaves dried at 450W power level, and the water activity was also obtained below 0.6, which was free from any bacterial spoilage during storage. A significant difference was found for all the dried mint samples in antioxidant activity, Vitamin C content, TPC, and chlorophyll content. Minimum Vitamin C degradation was observed in 450W dried samples, and high antioxidant was found in the mint leaves dried at 630W. Microwave vacuum drying can be considered the better drying method, mainly for leafy vegetables, which produce a final product with better retention of bioactive compounds.

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