Effect of microwave vacuum drying on nutritional composition of moringa (Moringa oleifera) leaves

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Abstract

Moringa (Moringa Oleifera) is well known for its medicinal and functional food properties. Microwave vacuum drying is an assisted drying technology that aims to preserve the nutrients and improve overall product quality. In this study, different microwave power levels and vacuum on the nutrients and phytochemical constituents were investigated. Moringa leaves were procured and subjected to microwave vacuum drying at three different microwave power levels (90W, 270W and 450W) at 500 mm Hg vacuum pressure. The nutrients of the leaves were determined using standard analytical methods. Data were analysed and interpreted using one-way ANOVA. There was no significant difference in the moisture and fat content between the dried samples. Crude fibre content tends to decrease with the increase in power level, whereas protein and total carbohydrate content tend to increase with the power level. Highest TPC and DPPH radical scavenging activity were reported at 450 W microwave power level. The total colour difference, i.e., was least in the sample dried at 450W and high for samples dried at 90W.

Keywords: Microwave, drying, nutritional, moringa, Moringa oleifera

Introduction

Moringa Oleifera belongs to the family moringaceae, and it is considered an extensively cultivated species of monogeneric family around the globe. Moringa Oleifera is native to the Indian subcontinent and is widely acclimated in tropical and subtropical regions, namely Africa, Asia, and South America (Mughal, Ali et al., 1999) [13] and (Somali, Bajneid 1984) [27]. Several synonyms also know moringa tree as 'The Spinach Tree,' 'Horse Radish Tree,' 'Drumstick Tree,' 'West Indian Ben,' (Ramachandran et al., 1980) [22], and in India, it goes by different names such as 'Murungai,' 'sahjan' or ‘sohanjana’ (Richter et al., 2003) [23]. Moringa Oleifera is labelled as the ‘miracle tree.’ It is a multipurpose and exceptionally nutritious vegetable tree with abundant potential uses. All parts of the tree, namely pod, fruit, leaves, are edible and a great nutrition source for all age groups. Moringa Oleifera leaves are considered to be a storehouse of rich nutritive compounds. It is a great source of digestible proteins, vitamins, minerals, and carbohydrates necessary for growth. The leaves are also free of anti-nutritive factors such as tannins and saponins (Fahay, 2005) [8]. Vitamin A and C, essential amino acids such as methionine, tryptophan, and lysine, are abundantly present in Moringa Oleifera leaves also consists of phytochemicals responsible for the antioxidant function. They also contain kaempferol and isoquercitrin as flavonoids.

The shelf life of fresh leaves (Moringa Oleifera) is a very short duration, approximately 2 to 3 days. Drying ensures reduction of water activity and improves the keeping quality of the leaves compared to the fresh leaves. The micro-nutrient content is even more in dried leaves in comparison with the fresh leaves. Common drying techniques employed for drying of M. Oleifera leaves include sun-drying and tray drying. Prolonged drying time, contamination of the product, loss of product quality accompanied by loss of nutritional value are some of the disadvantages involved when leaves are dried using conventional methods. The micro-nutrient content is even more in dried leaves (Mishra et al., 2012) [12]. Microwave drying can be employed to dry M. Oleifera leaves, higher thermal conductivity, precise control of the system, and energy-saving operations ensure that microwave drying has the upper hand in drying M. Oleifera leaves when compared to conventional methods (Potisate & Phoungchandang, 2015) [19].

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However, a rapid mass transfer would result in textural damage in some cases. Also, hot spots are formed due to the electric field's non-uniform distribution (Hu et al., 2006) [10]. To combat microwave drying limitation, microwave-assisted vacuum drying has been employed for drying fruits and vegetables. This method of drying rate of mass transfer is increased by increasing the pressure gradient between inner and outer layers and carrying out the drying process at low temperatures (Péré & Rodier, 2002) [18]. In the present context, the microwave-vacuum drying method can be considered as a novel drying technique. It allows shorter drying time and a considerable improvement in the quality of dried materials (Bondaruk et al., 2007) [4]. Microwave Vacuum drying is a combination of drying Technology where the potential of microwave energy is combined with a vacuum environment for rapid, low-temperature dehydration. Limitations of conventional drying techniques such as hot air drying can be addressed using microwave vacuum drying. Dried moringa leaves have great market potential. The quality characteristics and retention of bioactive compounds in moringa leaves are solely dependent on drying methods. Microwave vacuum drying technique can be employed in the drying of moringa leaves for better product quality and resulting in higher retention of bioactive compounds. This study aims to examine the nutritional value and phytochemical constituents of methanol extracts from present in the microwave vacuum dried moringa leaf powder and also study the impact of microwave vacuum drying on the quality parameters such as colour and water activity which helps in describing the fact that better quality can be achieved using this drying technique.

Material and Methods

Raw material

Fresh moringa leaves were procured from the local market at Thanjavur, Tamil Nadu. Diseased and damaged leaves were discarded manually, then leaves were washed in running tap water until the dirt is completely removed. Later, the washed leaves were soaked in 0.1% NaCl for 5 minutes to eliminate surficial microbes and leaves were further washed with distilled water. Excess water was removed by spreading leaves on a tray for a brief period (approximately 30 min) at ambient temperature. The leaves were then packed airtight and stored in refrigerated conditions (approximately 4 °C), which was used for further analysis.

Experimental setup

The experimental setup consisted of a domestic microwave oven (Model: IFB 30SC4) with a maximum magnetron output power of 0.9 kW of frequency-2450MHz, a glass vacuum desiccator, vacuum pump (Value double stage vacuum pump (VE215 N)), hose pipe, vacuum gauge, pressure regulator, and air drying unit. The glass vacuum desiccator containing the sample was put inside the microwave, and this was attached to the vacuum pump through a hose pipe. The pressure regulator was used to control the vacuum pressure within the system, and the vacuum gauge indicated the vacuum pressure held up in the glass desiccator. The air drying unit consisted of a conical vacuum flask with silica gel. This unit entrapped the water vapour due to the drying of the product and ensured that it maintains the vacuum level. Moringa leaves were dried at three different microwave power levels 90W, 270W and 450 W microwave power level, and the drying system was maintained at a vacuum level of 500 mm Hg. The dried moringa leaves were powdered using a food processor, and then the powder is packed in LDPE plastic pouches and stored in a desiccator. The dried moringa leaf powder sample is analyzed to determine the physical parameters (colour and water activity) and chemical parameters (moisture content, crude protein, crude fibre, crude fat, total phenols, and antioxidant activity). The phenolic and antioxidant activity profiles are determined using chromatographic methods.

Hunter color lab flex meter (Make: Hunter Association Laboratory, Inc., USA) is used to determine the color of a dried moringa leaves powder. The L*, a* and b* value indicates red to green scale (+a to –a), and b* value ranges from yellow to (+b to -b). ΔE is calculated as follows (Naik et al., 2020.)

$$\Delta E^* = \sqrt{(L - L^*)^2 + (a - a^*)^2 + (b - b^*)^2}$$  \(\text{(1)}\)

where $L^*$, $a^*$ and $b^*$ are the colour parameters of the Fresh moringa leaf sample and $L$, $a$ and $b$ are the color parameters of dried moringa leaves powder.

Water activity

The Water activity ($a_w$) of the dried tomato slices are determined using the Aqua Lab Water Activity Meter (4TE). This works on the principle of focusing the infrared beam on a tiny mirror, which determines the accurate dew point temperature and then translated into water activity. Water activity can be measured by placing a sample in a disposable cup in the water activity meter, sealing the sample chamber lid over the sample, and waiting for vapour equilibrium.

Proximate composition

Determination of moisture content

The moisture content of the samples was determined using the AOAC method (2005) [1]. 5g of the sample was then placed in a pre-weighed Petri dish and then placed in an oven to dry at 105 °C for five hours. The moisture dishes were cooled, and the final weight of the dish was noted.

$$\text{moisture content} = \frac{\text{Initial weight of sample (g) - Final weight of sample(g)}}{\text{Initial weight of sample (g)}}$$  \(\text{(2)}\)

Determination of crude fat

Crude fat was determined using the Soxhlet method (AOAC, 2005) [1]. 2g of sample was weighed and wrapped in filter paper, kept in a thimble. Sample contained thimbles were kept in an oil flask then 80ml of hexane solvent was added. Then oil flasks were kept in Soxhlet apparatus for processing. After completion of evaporation of solvent extract, contained flasks were dried at 105 °C for 30min. The final weight of the flask was noted.
**Determination of ash content**
The ash content was determined using the method described in AOAC (1995). 3g of the sample was weighed into a crucible in a muffle furnace and heated at 550 °C for six hours until it became greyish coloured ash. The crucible was removed from the muffle furnace with a tong’s help and placed in a desiccator for allowing it to cool. After cooling, it was reweighed, and the difference obtained the weight of the ash.

% Ash content = \( \frac{\text{weight of ash content (g)}}{\text{weight of sample (g)}} \times 100 \) …(4)

**Determination of protein**
Protein was analyzed by the Kjeldahl method (AOAC, 2005) \(^{1}\). 0.2 g of samples were weighed into the digestion tube, and 5g Na₂SO₄, 1g of CuSO₄ and 10mL of Conc H₂SO₄ are weighed into the digestion tube. The tubes were kept for digestion for three h. The digested tubes are further distilled with 40% NaOH, 4% Boric acid. The distillate is collected in a conical flask and titrated against 0.1N HCl with mixed indicator. Titrated value is noted when the solution turns pink.

% Nitrogen content = \( \frac{(TV−BV)\times N\times14.007\times100}{\text{weight of sample (g)}} \) …(5)

TV= Titrant volume of sample (ml), BV= Titrant volume of blank (ml), N= Normality of HCl

% Protein content = % nitrogen content × 6.25 …(6)

**Determination of crude fibre**
Crude Fibre content was determined by Weende's method (AOAC, 2005) \(^{1}\). 2g of the sample was weighed into a beaker, and 200mL of 1.25% H₂SO₄ was added, and the mixture was boiled for 30minutes. The solution was filtered with whatman filter paper; Then the residue was transferred into a beaker, and 200mL of 1.25% NaOH was added and boiled for 30 minutes, after which it was filtered and rinsed with distilled water. The residue was transferred into a crucible and placed in a hot air oven at 100 °C for eight hours to dry. Then the crucibles were removed and placed in a desiccator to cool before weighing. After weighing, the sample was incinerated, cooled in a desiccator, and reweighed.

**Determination of carbohydrate**
The carbohydrate content of the test sample was determined by estimation using the arithmetic difference method.

%CHO = 100 - (% fat + % ash + % fiber + % protein) …(7)

**Determination of energy content**
The energy values (kcal/100 g) were determined by multiply the values of carbohydrates, lipids and protein by a factor of 3.75, 9 and 4, respectively, and taking the sum expressed in kilocalories (Castillo-Lopez et al., 2017) \(^{15}\).

**Preparation of Methanol extracts for phytochemical analysis.**
Homogenizer assisted extraction was carried out to extract the polyphenol compounds from *Moringa Oleifera* leaves. 10 mL of methanol was added to one gram of dried sample, and extraction was carried out using an IKA (T18 ULTRA TURRAX) Homogenizer at 20,000 rpm for 3 minutes. The extracts were then centrifuged using REMI Centrifuge (C-24 Plus) at 10,000 rpm for 15 minutes at four °C. The extracts were subjected to concentration using a rotary evaporator. Later the methanol extracts were dried to total dryness, and 2mL of methanol was added for resuspension. The extract solution was stored in vials and refrigerated conditions for further analysis (Rocchetti et al., 2019) \(^{23}\).

**Determination of Total Phenolic Content**
The concentration of total phenolic content in the extracts was measured by UV spectrophotometer (SICAN 2301) based on colorimetric oxidation or reduction. Folin–Ciocalteau Reagent (FCR), an oxidizing agent, was used to estimate the phenolic compounds in the leaves' extracts. In a series of test tubes, 0.1 mL of the extract in methanol was taken, 0.9 mL of distilled water, mixed with 2 mL of FCR and 1 mL of sodium carbonate (20% w/v). The tubes were then allowed to stand in the dark at room temperature for one hour before the absorbance was read at wave-length set at 760 nm against a blank. A standard curve was prepared using Gallic acid monohydrate. Using the standard curve, the total phenolic compound content was calculated and expressed as Gallic acid equivalent (GAE) in mg/g of extracts (Sreelatha & Padma, 2009) \(^{28}\).

**Determination of DPPH radical scavenging activity**
The methanol leaf extracts' antioxidant activity was estimated by measuring the ability to scavenge 2,2'-diphenyl-1-picrylhydrazyl stable radical (DPPH). The spectrophotometrical assay was carried out as described previously (Nouman et al., 2016) \(^{15}\) with slight modifications. 0.5 mL solution of the extract was added to 0.004% DPPH solution in methanol. The contents were vortexed vigorously and allowed to stand in the dark at ambient temperature conditions for 60 min. A blank solution of methanol and DPPH was also prepared but without the sample. After one-hour incubation in the dark, the samples' absorbance and blank were read at 517 nm using a spectrophotometer (SICAN 2301). The capability to scavenge the DPPH radical was calculated using the equation.

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\text{DPPH radical scavenging activity} = \frac{\text{Absorbance control} − \text{Absorbance sample}}{\text{Absorbance control}} \times 100
\] (8)
replacement of magnesium compounds present in the chlorophyll by hydrogen molecules (Rudra et al., 2008) [20]. Slower degradation of chlorophyll occurs when subjected to a shorter drying process (Drouzas & Schubert, 1996) [7]. (Bondaruk et al., 2007) [4] reported that improvement of the color of potato cubes is found with decreasing pressure. The results obtained correlated with the microwave vacuum drying of mint leaves (Therdthai & Zhou, 2009) [29].

Fig 2: Colour degradation during microwave vacuum drying

Water activity
The water activity (aw) of the fresh sample and dried *moringa oleifera* leaf powder was 0.4171 to 0.4272. There was no significant difference in the water activity level at varied microwave power levels presented in Table 1. As the moringa's water activity values were lower than 0.60, they are considered safe and shelf-stable to microbial growth. (Rajkumar et al., 2007) [20].

Proximate composition
The determination of dried moringa powder's proximate composition is crucial since it is an assessing factor to ascertain its nutritional significance. The chemical composition of microwave vacuum dried moringa leaves powder samples dried at varying power levels of microwave wattage to determine moisture content, ash content, crude protein, fat, total carbohydrates and crude fiber, which are presented in Table 1. Generally, the nutritional composition increased as the leaves were subjected to drying. This could be the result of an increase in the internal composition of nutrient as moisture is reduced (Foline et al., 2011) [9].

Moisture content
The moisture content of the microwave vacuum dried moringa leaves powder ranged from 3.885±0.045% to 3.975±0.005%. There was no significant difference (p > 0.05) in the moisture levels of the samples dried under varied microwave power levels (90 W to 450 W). Rajput et al. (2017) [21] reported that fresh moringa leaves' moisture content was around 70-75 percent (w.b). Rigueiro-Rodríguez et al. (2002) [24] reported that for herbs safe moisture content for successful preservation is less than 15 percent. Ali, Yusof, Chin, & Ibrahim (2017) [2] reported that microwave dried moringa leaves' moisture content is about 4.93% (w.b), the results obtained in the agreement of the reported value. Microwave vacuum drying involves greater moisture loss because of the rapid removal of water-bound within the food matrix, which is greatly influenced by the combination of both microwave power and vacuum conditions. The moisture content of microwave vacuum dried garlic slices was reduced to less than 5% (w.b) rapidly due to the microwave and vacuum's combined effect. (Wang et al., 2011) [20].

Crude fat
Fat content ranged from 7.214±0.133% to 7.296±0.050%. There was no significant difference between the crude fat level at varied microwave power levels (p > 0.05). The results obtained agreed to the crude fat content reported by (Istiau et al., 2015) [11]. Microwave drying may result in fat oxidation and fatty acid isomer formation (Bashir et al., 2020) [3]. Vacuum conditions in this particular study would influence the reported fat content (Panyakamma et al., 2019) [17].

Ash content
The ash content ranged from 6.393±0.073% to 7.524±0.0878%. There was a significant difference in ash contents (p < 0.05). The ash of the dried leaf powder is considered the measure of mineral content of the original food product. The ash content of the present study was in good agreement with the ash content reported by Istiau et al., 2015) [11] around 11.5%.

Protein
Dried moringa leaves could serve as a great protein source in human diets. There was a significant difference (p < 0.05) in the protein content between the samples dried at 90W (25.296±0.495) and samples dried at 450 W (31.230±0.883), there was no significant difference between the protein content of 270W (28.816±0.0851) dried samples and 450W (31.230±0.883) dried samples in this particular study (p > 0.05). Ali et al. (2017) [2] recorded 29% of crude protein in microwave dried moringa leaves. This result was close to the value obtained in the current study. The decrease in protein levels with decrease power levels and prolonged drying time may be attributed to a browning reaction involving loss of protein (Bashir et al., 2020) [1].

Crude fibre
A significant difference (p < 0.05) in crude fibre level was observed among the samples. The highest fibre content was observed in samples dried at 90W (15.228±0.238) and the lowest crude fibre content in samples dried at 450W (11.926±0.189). Oblade, Akanbi, Olunlade, & Adeola (2015) [16] reported that crude fibre content for oven-dried moringa leaves was around 17.56% to 17.26%, the reported values in this study is in agreement with the value obtained in this particular study.

Carbohydrates and Energy content
The carbohydrate reported was highest in samples dried at 90 W (41.810±0.668) and lowest in samples dried at 450 W (38.209±0.619), there was a significant difference (p < 0.05) in carbohydrate levels between the samples subjected drying at 90W and 450 W. There was no significant difference between the samples dried at 270 W (40.391±0.758) and 450W (38.209±0.619), Oblade et al. (2015) [16] reported the carbohydrate to be around 45.55% to 35.99% for moringa leaves dried under varied oven temperatures. When subjected to high power levels, the decrease in carbohydrate content can be attributed to sugar's rapid caramelization at the higher temperature. When subjected to heating carbohydrates having less molecular weight is lost. The energy content ranged from 323.647±0.037 kcal/kg to 333.229±0.756 kcal/kg. There was a significant difference in the energy content. The energy content of microwave dried moringa leaves was 380 kcal/kg.
(Ali et al., 2017) [2]. The results obtained in this particular study was close to the reported values.

**Total Phenol Content and DPPH Radical scavenging activity**

The antioxidant in plants is mainly due to the Total phenolic content since it acts as the main contributor to the antioxidant activity in herbs (Siti Mahirah et al., 2018) [20]. Total phenolic content (TPC) was determined from the calibration curves of Gallic acid. Antioxidant activity is reported in terms of DPPH radical scavenging activity. It was found that all the dried samples showed a significant increase in TPC, DPPH radical scavenging activity. TPC and antioxidant activity are reported higher in the sample dried at 450 W and 500mm Hg pressure.

| Parameters          | 90W   | 270W   | 450W   |
|---------------------|-------|--------|--------|
| Moisture (%)        | 3.97±0.005<sup>a</sup> | 3.90±0.035<sup>a</sup> | 3.88±0.045<sup>a</sup> |
| Fat (%)             | 7.22±0.017<sup>a</sup> | 7.21±0.133<sup>a</sup> | 7.29±0.050<sup>a</sup> |
| Ash (%)             | 6.39±0.073<sup>c</sup> | 6.89±0.079<sup>b</sup> | 7.52±0.087<sup>b</sup> |
| Crude protein (%)   | 25.29±0.495<sup>a</sup> | 28.81±0.085<sup>b</sup> | 31.23±0.883<sup>b</sup> |
| Crude fibre (%)     | 15.22±0.328<sup>a</sup> | 12.77±0.197<sup>c</sup> | 11.92±0.189<sup>c</sup> |
| Total carbohydrate | 41.81±0.668<sup>b</sup> | 40.39±0.758<sup>c</sup> | 38.20±0.619<sup>c</sup> |
| Energy content (kcal/kg) | 323.647±0.037<sup>b</sup> | 331.663±1.767<sup>b</sup> | 333.229±0.756<sup>b</sup> |
| Total Phenolic content (mg/g dried leaves) | 11.35±0.661<sup>c</sup> | 14.00±0.198<sup>b</sup> | 16.40±0.240<sup>b</sup> |
| DPPH Radical scavenging activity (%) | 40.53±0.946<sup>c</sup> | 46.41±1.028<sup>c</sup> | 52.58±1.174<sup>c</sup> |
| Water activity (a<sub>ω</sub>) | 0.42±0.002<sup>a</sup> | 0.40±0.040<sup>a</sup> | 0.41±0.01<sup>a</sup> |

<sup>*Values are triplicate and represent in Mean ± SD; different superscripts in the same column mean that values are significantly different (p<0.05)</sup>

### Conclusion

This study aimed to analyze the nutritional content and physical parameters of microwave vacuum dried moringa leaf powder dried using a lab-scale microwave vacuum drier at varied microwave power levels. The Total colour difference, i.e., ΔE, was lower for samples dried at 450 W microwave power. The water activity of the dried moringa leaves was reported to be below 0.6. There was no significant difference in moisture content and fat content in the samples dried at varying power levels. A significant difference in crude fibre content, ash content, TPC and DPPH radical scavenging activity was found between all the samples. There was no significant difference in crude protein, carbohydrate and the energy content between the samples dried at a 270 W and 450 W microwave power level. Microwave vacuum drying is an assisted technology that results in superior product quality with minimal loss in nutritional and phytochemical aspects.

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