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The Impacts of Drying Methods on the Total Flavonoids and Antioxidant Capacity of Moringa Oleifera Leaves

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***Abstract*: *Moringa oleifera* is a versatile plant that has nutritional, medicinal, and socio-economic values. M. oleifera leaves are rich in minerals, vitamins, and other essential phytochemicals. Washing and drying is an important process in the M. oleifera leave processing industry for food and medicine. This study aims to analyze the impact of the leaves drying methods on their total flavonoid and antioxidant capacity. The four drying methods (oven-drying, room temperature-drying, sun-drying, and roasted-drying) were performed. Total flavonoid of the dried leaves were analyzed by spectrophotometric UV-Vis assays and antioxidant capacity was analyzed by DPPH (2.2-diphenyl-1-Picrylhydrazil) radical scavenging activity assay. The result showed that oven drying was the best method resulting in total flavonoids and antioxidant capacity of 12.84 mg quercetine equivalent/g (mgQE/g) and 66% respectively. Otherwise, the roasting-drying method results in the lowest total of flavonoids and antioxidant capacity, probably because of excessively high and unstable temperatures. Heat-sensitive phytochemicals were degraded or bio transformed at high temperatures. Based on this research that the drying method can be affect the level of flavonoid content and antioxidant.**

***Keywords*: Antioxidant, Moringa oleifera, Drying method, Flavonoid contents.**

1. Introduction

Moringa oleifera, is a member of the Moringaceae family of perennial angiosperm plants [1]. Moringa oleifera has been popular for several centuries as a multipurpose plant, abundant nutrition and has medicinal properties. The plant is also known as Horse - radish tree, Drumstick tree [2]. Every part of this plant contains a valuable medicinal feature Moringa is rich in nutrition presence of a variety of essential phytochemicals present in its leaves, pods and seeds [3] The leaves can be consumed in raw, cooked or dried and ground in to powder that can be added to any food as a nutrient supplement [4]. One hundred grams of dry Moringa oleifera leaf contains 9 times the vitamin A of carrots, 15 times the potassium of bananas, 17 times the calcium of milk, 12 times the vitamin C of oranges and 25 times the iron of spinach. Antioxidants galore and plant leaves of munga contains rich source of antioxidants, including beta carotene, vitamin c, quercetin and chlorogenic acid [5]. There are several of Moringa oleifera leaf processing which one of them is drying process [6]. Drying is the process to decreasing moisture content in the feed hereby inhibiting microbial and enzymatic reactions allowing to be preserved. People are used to dry some leafy plant material in order to long-term preservation and risk of contamination. Drying is an effective method that increases the shelf life of the final product by slowing the growth of microorganisms and preventing certain biochemical reactions that may alter their characteristics. Drying temperature has been identified as an important factor because there are changes in the nutritional composition that are effected by different drying temperatures [7] Basically, drying is divided into two types, natural drying and artificial drying. Natural drying used sunlight while artificial drying uses the instrument and can be manipulated by human [8]. In the processing should pay attention to the quality of the leaves Moringa oleifera, because the product will be consumed [9]. Therefore, this research conducted to analyze the impact of the leaves drying methods on their total flavonoid and antioxidant capacity. The four drying methods (oven-drying, room temperature-drying, sun-drying, and roasting) were performed.

2. Material and Methods

A. Drying Methods

Moringa oleifera leaves washed using well water then soaked in 1% salt solution, the salt used are industrial salt and raw salt. After washing the drying process with several methods (oven-drying, room temperature-drying, sun-drying, and roasting). Drying with oven method is done at 40 ° C for 24 hours. Roasting by using a heated frying pan then the leaves are roasting until dry (can be kneaded). Drying with sunlight for 3 days from 08.00 until 14.00 until the leaves are dry and fragile. Room drying, Moringa oleifera is placed on trays in a room with good ventilation conditions, this drying for 4 days.

B. Moisture Content

The moisture content is determined based on the method of [10]. Leaf samples that have been given a drying treatment are weighed as much as 2 g, then reheated by oven at a temperature of 105 ºC for 3 hours, then cooled for 30 minutes, then weighed.

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Determination of moisture content is calculated by the:

$$\text{Moisture Content} = \frac{A - B}{A}\ 100\%$$

Information:
A = sample weight before drying
B = sample weight before drying

C. Extraction Procedure

Extracts were made based on [11] with modifications. 10 g of dried *Moringa oleifera* leaf sample was crushed and dissolved in 200 ml of 70% ethanol, then soaked for 3 days while occasionally shaking. After 3 days, the sample homogenized using a centrifuge at 8000 rpm for 10 minutes, the supernatant was taken and evaporated using hot plate until the extract thickened.

D. Total Flavonoid

To determine the total flavonoids, a quercetin curve is needed by weighing as much as 25 mg of quercetin and dissolved in 25 ml of ethanol. Furthermore, 1 ml of the stock solution was pipetted and the volume was made up to 10 ml with ethanol to obtain a concentration of 100 ppm. From a standard solution of 100 ppm quercetin, some concentrations were made, 20 ppm, 40 ppm, 60 ppm. From each concentration of the standard solution of quecetin, 1 ml was pipetted then 1 ml of 10% AlCl3 and 1 ml of potassium acetate were added. After that the samples were incubated for 1 hour at room temperature. The absorbance was determined using the UV-Vis spectrophotometric at 435 nm [12].

Measurement of total flavonoids based on [13]. Weighed 100 mg of extract, then dissolved in 10 ml ethanol, then the extract was taken 1 ml and added 3 ml of ethanol on the measuring flask. Then, 0.2 ml AlCl3 10% and added 0.2 ml of potassium acetate, chopped with aquades up to 10 ml. The solution is stored for 30 minutes in a dark place at room temperature. The absorbance is determined using the Uv-Vis spectrophotometry method at a wavelength of 435 nm. Determination of flavonoids values is done based on formula [14]:

$$\text{Total Flavonoid} = \frac{C \times V}{W}$$

C = Flavonoid concentrations that calculated using standard curve equations
V = volume of solvent (ml)
W = weight of powder (g)

E. Antioxidant Capacity

Determination of antioxidant capacity based on Shimamura [15] and modified. *Moringa oleifera* leaf extract was taken as much as 300 µl and then added 900 µl of DPPH solution (2.2-diphenyl-1-Picrylhydrazil) was then homogenized using a vortex. Furthermore, the solution is incubated for 30 minutes at room temperature in the dark place. Then absorption of mixture is measured at 517 nm wavelength with UV-vis spectrophotometer. 300 µl Ethanol 70% plus 900 µl DPPH is used as Blanko. The percentage of antioxidant activity can be calculated using the following formulas:

$$\%\ \text{inhibisi} = \frac{\text{A blanko} - \text{A sample}}{\text{A blanko}}\ \text{x}\ 100\ \%$$

3. Results

A. Moisture Content

Based on Figure 1 the highest moisture content is room drying, with 8% and the lowest is in the roasting method.

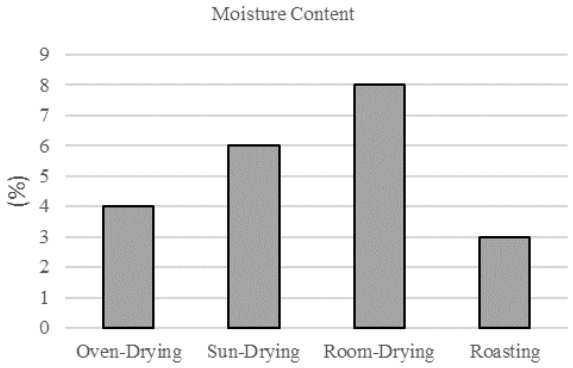

Fig. 1. Moisture content of some drying methods

The decrease in the value of the moisture content of *Moringa oleifera* leaves is influenced by the drying temperature and drying time. According to Winarno [16], the process of evaporation of water will occur quickly due to the high drying temperature, which causes the water content in the material to be low. And if there is an increase in the temperature, the material that has the ability to release water from its surface will release more water [17].

B. Total Flavonoid

Based on the Figure 2 can be shown that the highest total flavonoids achieved by oven method with total flavonoids of 12.84 mgQE/g while the lowest total flavonoids obtained Roasting method of 3.90 mgQE/g. Oven method has a higher total of flavonoids can controlled temperatures resulting in the stability of the flavonoids compounds.

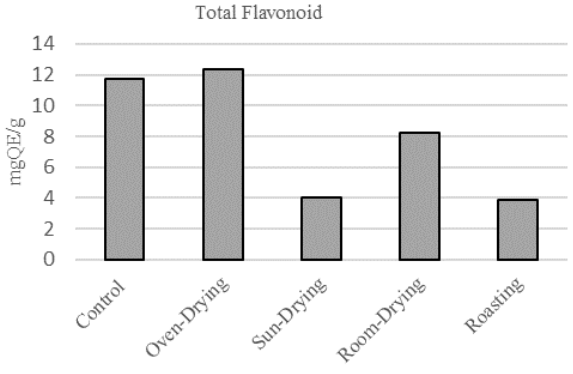

Fig. 2. Analysis of total flavonoid

Instead, drying with the roasting method obtained the lowest a total of flavonoids and antioxidant capacity can cause from excessively high and unstable temperatures. Drying can damage the phytochemical substances by thermal breakdown

that affect the integrity of the cell structure which then resulted in the migration of components, leading to losses by leakage or breakdown by various chemical reactions involving enzymes, light and oxygen. The loss of flavonoids during drying might be due to the process conditions, in some temperatures and the duration of drying [18].

C. Antioxidant Capacity

The antioxidant capacity dried *Moringa oleifera* leaves was determined by DPPH methodologies. DPPH assay is simple and one of the most widely used methods. DPPH has been widely used to evaluate the free radical scavenging activity of various antioxidant substances. DPPH is a stable free radical which gives rise to the deep violet color in methanol solution, the remaining violet DPPH radical is measured by a UV-Vis spectrophotometer at approximately 515–520 nm. In the DPPH assay, the antioxidants which can donate hydrogen are able to reduce stable DPPH radical to the yellow colored, non-radical form of DPPH-H [19], [20]. Antioxidant from natural source can improve the antioxidant system in body for scavenging free radicals. An interest in antioxidant from natural sources increasing faster than synthetic sources. Phenolic compounds which naturally present in M. oleifera plant can reduce the risk of many diseases and its effects which correlated with the antioxidant compounds. Recently, there are some reports about M. oleifera leaves which rich in phenolic compounds such as flavonoids, gallic acid, and quercetin as antioxidant capacity [21].

Based on the Figure 3 can be shown the highest of antioxidant capacity achieved by oven drying of 66% and the lowest antioxidant capacity obtained roasting method. Antioxidant capacity of *Moringa oleifera* leaves may be related to the amount of total flavonoids. Since these compounds act as scavengers of the free radicals produced during oxidation reactions. In order to explore the influence of the phytochemical compounds on *Moringa oleifera* leaves antioxidants capacity. Drying processes, and in particular high temperature and long drying time, might destroy some of the biochemical compounds [22] and promote a decrease the antioxidant capacity [20].

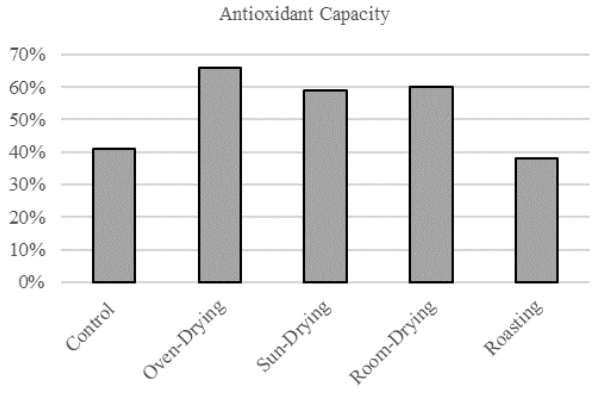

Fig. 3. Analysis of antioxidant capacity

In control or (fresh sample) has relatively low antioxidant capacity revealed to the dried leaves of *Moringa oleifera*, it is in accordance with the statement Hossain et al [23] the samples are largely prone to internal and external stresses, such as enzymatic degradation and oxidation by atmospheric oxygen, respectively. Increased antioxidant capacity in the drying process can be caused by low water content because the drying process will cause substances contained in food ingredients become more concentrated [8].

4. Discussion

The research finding presented, the effects of four drying methods, oven drying, sun drying, room drying and roasted on the total flavonoid and antioxidant capacity of the leaves were investigated. It was noted that the oven drying is the best method among the other three methods for conserving the total flavonoid and antioxidant of leaves that was consistent with earlier study. Analyzing the results of drying methods on the whole flavonoid capacity, it was found that oven drying method was the most suitable for keeping the total phenolics. [24] showed that the oven-dried red seaweed (Kappaphycus alvarezii 'crocodile' morphotype) samples that contain higher of total phenolic contents compared with other drying methods. On, [25] research result that drying gave increase of the whole phenolic capacity in fruits and vegetables. However, oven drying was found to result in a decrease of phenolic in Neem leaves as it was compared to shade drying (Sejali and Anuar, 2011) the oven-dried leaf samples appeared to have the highest level of total flavonoid contents (301.20 mg GAE/100 g DW). [26] study reported that oven drying gave increase of DPPH scavenging activity of Iranian quince in comparison with sun drying. But based on another research [27] the antioxidant activity was high loss in oven drying than cabinet dryer. Intense thermal process also might cause significant loss in antioxidant, it showed by the lowest antioxidant activity was 28.05 % ± 1.54 % at oven drying 60 ºC. That was naturally in plants as well as deactivate enzymes and degrade phytochemicals [28] the decreasing of antioxidant activities has also been correlated to Maillard- type antioxidants declined generation and accumulation. Several studies reported related of antioxidant with phenolic and flavonoid content. The research in Rhus flexicaulis Baker described that high antioxidant activity might be attributed to the high phenolic and flavonoid content [29] this is caused by during the drying process, loss of macromolecules such as polyphenols occurred, which was associated with the temperature and the length of time [30].

5. Conclusion

Based on the research that has been done can be conclude all drying methods (oven-drying, room temperature-drying, sun-drying, and roasting) had adverse effects on total flavonoids and antioxidant capacity of Moringa oleifera leaves. Drying by oven method has the best level of hygiene and quality compared to other drying methods, with a total of flavonoids 12.29 mgQE/g and antioxidant capacity of 66%. While the roasting drying method has the lowest quality with a total of 3.90 mgQE/g flavonoids and antioxidant capacity 38%. Further research needs to be done using more controlled temperatures.

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