Evaluation of nutrient and antioxidant activity on steam blanching of *Moringa oleifera* leaves

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Abstract. *Moringa oleifera* is a plant that has high nutritional content. Post-harvest treatment take effect in improving product quality. This study was carried out to evaluate nutrient and antioxidant activity of steam blanched moringa oleifera leaves. Sample of moringa oleifera were prepared where the temperature 80°C in ±5 minutes for the steam blanched. The fresh *M. oleifera* and steam blanched *M. oleifera* leaves dried at 60°C were analyzed for their proximate, mineral, total phenol, total flavonoid, and antioxidant activity contents. The analysis revealed that steam blanched leaves contained more dry matter, crude protein, crude fat, and phenolic total, and flavonoid than unblanched leaves. Therefore, its less fibre and mineral. This study showed steam blanching can serve as alternative post-harvest treatment to increase nutritional value of moringa oleifera leaves.

1 Introduction

*Moringa oleifera* currently receive attention because of its benefits and good nutritional content. *M. oleifera* is a plant that can grow in tropical and subtropical areas so that it has been cultivated in various countries [1]. Parts of moringa oleifera such as leaves, stems, flowers, and roots have good nutritional content and can be used as medicine [2,3,4]. Leaves, flowers, and pods used by people as vegetables, otherwise its can be used as feed for livestock [5,6].

*M. oleifera* tree is rich source of certain macro and micronutrient also its good biomass production [7,8]. High biomass production effect of fast growing from *M. oleifera* tree [9]. The *M. oleifera* plant exhibit compound of zeatin, quercetin, β-sitosterol, caffeoylquinic acid, and kaempferol [5]. *M. oleifera* leaves contain highly nutritional value than other part [10]. *M. oleifera* leaves have been analyzed to be high source of β-carotene, vitamin especially vitamin C, protein, and mineral also act as source of natural antioxidant. As high source of mineral it contains high concentration of zinc, magnesium, calcium, potassium, phosphor, iron, manganese, and copper [7]. It is reported as a good source of dietary fibers; proteins; vitamins; minerals; lipids; and water [11]. In fact, the leaves of *M. oleifera* contain more calcium and twice as much protein than milk, higher vitamin C than oranges, higher

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vitamin A than carrots, and higher potassium and iron than bananas [12]. *M. oleifera* leaves are unique because of their remarkable source of minerals but lower amounts of antinutritional compounds [6]. The leaves of *M. oleifera* have also been reported have numerous phytochemical compounds [13]. The *M. oleifera* leaves extract contain high active compounds such as polyphenol and flavonoid also antioxidant capacity [14,15]. Its useful properties of *M. oleifera* leaves [16]. Antioxidant activity on *M. oleifera* leaves have potential against free radicals and protect from oxidative damage [17].

Further it has been shown that *M. oleifera* is useful for humans and livestock. *M. oleifera* can contribute meeting human and livestock nutritional requirement as multipurpose plant [18]. As traditional medicine *M. oleifera* widely used in human [15]. *M. oleifera* leaves have potential as treatment or prevention of cardiovascular disease and diabetes [4]. The active compounds of *M. oleifera* can active against hepatocarcinoma and leukemia cell, its indicated *M. oleifera* as anticancer [19,20]. Furthermore, *M. oleifera* has potential application in livestock production and products [21]. The nutrition of *M. oleifera* leaves are necessary weight gaining and milk production of livestock [22]. Inclusion *M. oleifera* leaf meal on poultry diets can increase economic benefits [5]. *M. oleifera* as feed supplement enhance the health status and sustain the production performance in poultry [23].

The development of post-harvest treatment at this time still needs to be concerned to improve product quality. Steam blanching can be beneficial to increase quality of products [24]. Also, thermal blanching contributes to the inactivation of various enzymes (polyphenol oxidase and peroxidase), soften texture, and destroy microorganism [25,26]. However, the effect of thermal processing on phytochemical contents can be dissimilar in different products [27]. Therefore, the objectives this study is to evaluate of nutrient and antioxidant activity on steam blanching of *M. oleifera* leaves.

## 2 Material and methods

### 2.1 Plant material and preparation

Fresh leaves of moringa oleifera were obtained from Sukaluyu village, Bogor, West Java Province, Indonesia. The leaves were divided into two pieces and one piece was steam blanched at 80°C for 5 minutes and the other was kept un-blanched. Steam blanching was done by artificial machine. The fresh and steamed leaves was dried in oven with lighting for heating at 60°C for 24 h. The dried leaves were powdered using electric blender and stored in air-tight container for further use.

### 2.2 Chemicals

The chemicals used for analysis are Ethanol 70%, folin ciocalteu, Natrium chloride, gallic acid, quercetin, alumunium chloride, 2,2-diphenyl-1-picrylhydrazyl (DPPH), aquadest, HNO₃, H₂SO₄, HClO₄, and HCl.

### 2.3 Nutritional determination

Fresh and steamed blanched *M. oleifera* dried powdered leaves were assessed for dry matter (DM), ash, crude protein (CP), crude fat, crude fibre (CF) using the Association of
Official Agricultural Chemists [28], mineral components were analyzed using the Atomic Absorption Spectroscopy (AAS) instrument [29,30].

2.4 Preparation of leaves extract for total phenolic, flavonoid, and antioxidant activity analysis

100 g of the dried leaves powder was soaked in 500 ml ethanol 70% in a conical flask and wrapped with aluminium foil for 24 hours with occasional shaking. After 24 hours, the extract was filtered using Whatman filter paper No: 1 repeated two times. The extract is separated from the solvent by a rotary evaporator. To remove a residual solvent, the extract is placed in the oven at 60 °C to obtain constant weight [31,32].

2.5 Determination of total phenolic

The content of total phenolic compounds was determined using Folin-Ciocalteu procedure [33].

2.6 Determination of flavonoid

The content of total flavonoids was analyzed using aluminium chloride colorimetric method [34].

2.7 Determination of antioxidant activity

Free radical scavenging activity was measured using DPPH (2, 2-diphenyl-1-picrylhydrazyl) method according to [35,36].

3 Results and discussion

3.1 Proximate analysis of *M. oleifera* leaves powder

Proximate compositions of fresh and steam blanched *M. oleifera* leaves powder are showed in table 1.

| Nutrients                | Fresh *M. oleifera* leaves powder | Steam blanched *M. oleifera* leaves powder |
|-------------------------|----------------------------------|------------------------------------------|
| Moisture (%)            | 10.46                            | 7.94                                     |
| Ash (%)                 | 8.89                             | 8.19                                     |
| Crude Protein (%)       | 30.17                            | 31.37                                    |
| Crude fat (%)           | 6.86                             | 8.35                                     |
| Crude fiber (%)         | 4.42                             | 3.47                                     |

The moisture of fresh *M. oleifera* leaves powder higher (10.46%) than steam blanched *M. oleifera* leaves powder (7.94%). It’s confirmed by the result of Rathnayake and Navarathna [37]. Prevention of microbial activity and increase in shelf life occurs if the moisture content is less than fifteen percent [24].

The steam blanched leaves had lower ash content (8.19%) than fresh leaves (8.89%). Ash in food largely determines the level of minerals that may be found in product [38]. The steam blanching could cause losing some of minerals [24,37].
The crude protein content on steam blanched \textit{M. oleifera} leaves higher (31.37\%) than that of fresh leaves powder. This result within agreement of reported by Moyo et al. [1], Rathnayake and Navarathna [37], and Mutiara et al. [39]. this makes \textit{m. oleifera} has the potential as a source of protein in animal feed. Cells damaged by steam blanching can reduce moisture thereby increasing protein content. Protein quality of \textit{M. oleifera} can be paired with meat and eggs, with its amino-acid composition corroborating with the FAO reference protein [40]. \textit{M. oleifera} leaves also contain all the essential amino acids [23,1].

The level of crude fat higher in the steam blanching (8.35\%) than fresh leaves (6.86\%). the amount of energy available determined by fat in food [38]. Therefore, the steam blanching had lower crude fiber (3.47\%) content than fresh leaves (4.42\%). this can be very useful as poultry that requires low-fiber feed. \textit{M. oleifera} leaf meal can be used as alternative feed for poultry [5].

\subsection*{3.2 Mineral compositions of steam blanched \textit{M. oleifera} leaves powder}

Data represented in Table 2 illustrates the mineral content of the two Moringa samples fresh and steam blanched leaves.

\begin{table}[ht]
\centering
\begin{tabular}{|l|l|l|}
\hline
Mineral & Fresh \textit{M. oleifera} leaves powder & Steam blanched \textit{M. oleifera} leaves powder \\
\hline
Ca (mg/100) & 2310.78 ± 4.29 & 2033.03 ± 4.17 \\
K (mg/100) & 1329.82 ± 23.82 & 1119.65 ± 14.92 \\
Na (mg/100) & 224.03 ± 0.16 & 138.08 ± 0.16 \\
Mg (mg/100) & 331.37 ± 0.37 & 326.11 ± 0.45 \\
P (mg/100) & 12.95 ± 0.26 & 12.39 ± 0.26 \\
Fe (ppm) & 152.89 ± 2.30 & 143.85 ± 3.78 \\
Zn (ppm) & 32.72 ± 0.14 & 28.45 ± 0.15 \\
Mn (ppm) & 25.44 ± 0.12 & 25.10 ± 0.09 \\
Cu (ppm) & 73.09 ± 0.12 & 72.71 ± 0.25 \\
Co (ppm) & 0.157 ± 0.02 & 0.133 ± 0.02 \\
\hline
\end{tabular}
\caption{Mineral compositions of fresh and steam blanched \textit{M. oleifera} leaves powder}
\end{table}

The minerals except Cu higher in fresh leaves than steam blanched leaves. The decrease in the minerals of the steam blanched leaves could be due to the draining of minerals in to the cooling water, from the cells which are damaged from condensed steam [41]. Variations in mineral content in the current study compared to previous studies can be related to variations in soil conditions, growing areas, plant varieties and maturity levels, etc [37]. This is an indication of a decrease in the ash content in the steam blanched leaves.

The Ca and K content obtained in this study is lower than that the values reported by Rathnayake and Navarathna [37] but higher in Fe content. Ca is considered as one of the important minerals for human growth and good dietary of Zn increase reproductive system [42]. Mn, Co, and Zn plays an important role in metabolism system [38]. Fe is an important component of Haemoglobin and Myoglobin for oxygen transport and cellular processes, deficiency of Fe leads to anaemia [37,38]. Ca/P ratio is an index of bone formation [43]. If Ca/P is low (low calcium, high phosphorus) more than the normal amount of Ca can be lost in the urine thereby lowering calcium levels in the bones [44].
3.3 Phytochemical contents and antioxidant activity of extracts of steam blanched *M. oleifera* leaves powder

Total phenolic, total flavonoid, and antioxidant activity content of fresh and steam blanched *M. oleifera* showed in Table 3. The result in Table 3 revealed that extract steam blanched leaves had higher total phenolic (348.26 ± 3.07), total flavonoid (5.32 ± 0.30) content, and IC50 value (105.36 ± 1.59) than extract fresh leaves.

Table 3. Phytochemical contents and antioxidant activity of extracts fresh and steam blanched *M. oleifera* leaves powder

| Parameters                  | Extract fresh *M. oleifera* leaves powder | Extract Steam blanched *M. oleifera* leaves powder |
|-----------------------------|-------------------------------------------|-------------------------------------------------|
| Total Phenolic (mg GAE/100 g) | 242.282 ± 2.31                             | 348.26 ± 3.07                                   |
| Total Flavonoid (mg QE/100 g)   | 5.26 ± 0.13                                | 5.32 ± 0.30                                     |
| EC50 value (ppm)             | 49.42 ± 1.12                               | 105.36 ± 1.59                                  |

GAE: Gallic acid equivalent, QE: Quercetin equivalent

Previous data for *Moringa oleifera* leaves were higher with those reported by [45] who quantified that the total phenolic content was 12.79 mg GAE/100g of *M. oleifera* leaves. Phenolic compounds are phytochemicals in plant. *M. oleifera* leaves contain two types of antioxidant compounds such as ascorbic acid, flavonoids, phenolics and carotenoids, it acts as a good source of natural antioxidants [46]. The higher component of total phenolic on steam blanched leaves due to ability of thermal blanching to inactivate enzymes such as polyphenol oxidase and peroxidase [25]. There are more than 8000 individual plant phenolic compounds and divided into two main groups as flavonoids and non-flavonoids [24].

The results of total flavonoid content were higher with recorded by Erian et al. [47], who found that total flavonoid contents in *M. oleifera* was 2.04 mg QE/100g. Flavonoids have long been acknowledged to possess anti-inflammatory, anti-allergic, antiviral, anti-proliferative and anti-carcinogenic activities [48]. Flavonoids and tannins are phenolic compounds and plant phenols are the main group of compounds that act as primary antioxidants or free radical scavengers [49].

The application of the current method is checked, in particular the use of parameters EC50 (substrate concentration to produce a 50% reduction of DPPH) [50]. The smaller the value EC50 means the stronger the antioxidant power [51]. Lower antioxidant capacity on steam blanched leaves due loss of vitamin C. High temperature on steam blanching process increase the risk loss of vitamin C [52]. Vitamin C is an excellent antioxidant and free radical scavenger, able to protect cells from oxidative damage by oxidants [14]. Vitamin C is easily destroyed by excessive heat and water, also exposure to air [53].

4 Conclusions

This study shows the steam blanched *M. oleifera* leaves, to be the most preferred in terms of phytochemical content. Steam blanching increase crude protein and crude fat content also decrease crude fibre content. Steam blanching decrease the minerals content. The high content of minerals is Ca followed K and Mg. Total phenolic and total flavonoid increase but decrease Antioxidant activity. Steam blanching can serve as alternative post-harvest treatment to increase nutritional value especially phytochemical content of moringa oleifera leaves.
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