

DETERMINATION OF MINERALS CONTENT IN LEAVES OF MORINGA OLEIFERA BY NEUTRON ACTIVATION ANALYSIS

PENENTUAN KANDUNGAN MINERAL DALAM DAUN KELOR DENGAN ANALISIS AKTIVASI NEUTRON

Th Rina Mulyaningsih, Saeful Yusuf
Pusat Teknologi Bahan Industri Nuklir- BATAN
e-mail : thrinam@batan.go.id

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ABSTRACT

DETERMINATION OF MINERALS CONTENT IN LEAVES OF MORINGA OLEIFERA BY NEUTRON ACTIVATION ANALYSIS. *Moringa oleifera* (Mo) or kelor is a high-value plant, distributed in many tropical and subtropical countries. This plant can be used as a vegetable, cosmetic oil, and medicinal plant. The leaves of Mo is rich in essential minerals needed by the body's health. *Moringa oleifera* widely cultivated in various places in Indonesia, easily obtained and inexpensive so precious can be used to overcome malnutrition. Research on the macro-micro mineral content in *Moringa* leaves here is still limited. Mineral composition in leaves include Mo leaves varies depending on location grow. The purpose of this research is to determine the mineral content in the leaves of the Mo taken from Indonesia. For the preliminary study samples was taken from Central Java to be compared with the results of studies conducted in several other countries. Leaf samples were collected randomly from sampling area. Mineral content in the samples is determined using Neutron Activation Analysis (NAA). Irradiation was carried out at rabbit system of Multipurpose Reactor G.A Siwabessy on neutron flux $\sim 10^{13} \text{ n.cm}^{-2}.\text{dt}^{-1}$. The results obtained indicate that Mo is rich in essential minerals, mainly Ca, Mg, K, Zn, Fe and Cl. Content in dried leaves include: calcium (3.45 %), magnesium (0.66 %), potassium (3.35 %), chloride (0.25%), iron (147.20 mg/kg), sodium (152.52 mg/kg), zinc (35.71 mg/kg), and manganese (102.10 mg/kg). Mo also contains other minerals such as chromium (4.76 mg/kg), bromine (4.82 mg/kg), cobalt (0.16 mg/kg), and aluminium (150.40 mg/kg) in addition to other element. Compared with the results of existing studies, it shows that mineral composition in Mo leaves varies depending on the location where the plant is grown.

Keywords: *Moringa oleifera*, leaf, mineral, essential, medicinal plant

ABSTRAK

PENENTUAN KANDUNGAN MINERAL DALAM DAUN KELOR DENGAN ANALISIS AKTIVASI NEUTRON. *Moringa oleifera* (Mo) atau kelor adalah tanaman yang bernilai tinggi, terdistribusi di banyak negara tropis dan subtropis. Tanaman ini dapat digunakan sebagai sayuran, minyak kosmetik, dan sebagai tanaman obat. Kelor kaya akan mineral penting yang dibutuhkan oleh tubuh, mudah diperoleh karena banyak dibudidayakan di Indonesia sehingga dapat digunakan untuk mengatasi malnutrisi. Penelitian tentang kandungan makro-mikro mineral dalam daun kelor di sini masih terbatas. Pada umumnya komposisi mineral di dalam tanaman termasuk kelor bervariasi tergantung lokasi tumbuh. Tujuan dari penelitian ini adalah menentukan kandungan mineral dalam daun kelor yang diambil dari Indonesia. Sebagai studi awal diambil sampel dari Jawa Tengah untuk dibandingkan dengan hasil studi yang dilakukan di beberapa negara lainnya. Sampel daun dikumpulkan secara acak dari daerah sampling. Kandungan mineral dalam sampel ditentukan dengan menggunakan analisis aktivasi neutron (AAN). Iradiasi dilakukan di fasilitas rabbit system Reaktor Serbaguna G.A Siwabessy pada fluks neutron $\sim 10^{13} \text{ n.cm}^{-2}.\text{dt}^{-1}$. Hasil yang didapat menunjukkan bahwa daun kelor kaya akan mineral penting, terutama Ca, Mg, K, Zn, Fe dan Cl. Kandungan dalam daun kering meliputi: kalsium (3,45%), magnesium (0,66%), kalium (3,35%), klorida (0,25%), besi (147,20 mg/kg), natrium (152,52 mg/kg), seng (35,71 mg/kg), dan mangan (102,10 mg/kg). Daun kelor juga mengandung mineral lain seperti kromium (4,76 mg/kg), brom (4,82 mg/kg), kobalt (0,16 mg/kg), dan aluminium (150 mg/kg), disamping unsur lainnya. Dibandingkan dengan hasil penelitian yang telah ada, ternyata komposisi mineral ini bervariasi tergantung pada lokasi dimana tanaman ini tumbuh.

Kata Kunci : *Moringa oleifera*, daun, mineral, esensial, tanaman obat

INTRODUCTION

Moringa oleifera (*Mo*) belongs to a family of Moringaceae. It is also commonly known as *kelor* in Indonesia. It is widely cultivated in Africa, Central and South America, Sri Lanka, India, Mexico, Malaysia, Indonesia and the Philippines. The tree is considered one of the world's most useful trees, as almost every part of the tree can be used for food, as an ingredient of Indian traditional medicine or has some other beneficial property [1]. This tree is known as the miracle tree or trees of life because it has rich nutrition and the many usages of this tree. *Mo* is used as a drug in treatment of many disease as cardiac and circulatory stimulants, posses antitumour, antipyretic, antiepileptic, anti-inflammatory, antiulcer, antispasmodic, antihypertensive, cholesterol lowering, antioxidant, antidiabetic, antibacterial and antifungal [2]. *Mo* leaves are highly nutritious, the young leaves are edible and are commonly cooked and eaten like spinach or used to make soups and salads. The most consuming part is the leaves because the needed nutrients are concentrated in the leaves [3].

Moringa oleifera leaves have been reported to be a valuable source of both macro and micro nutrients, that needed by the human body, being a significant source of beta-carotene, vitamin C, protein, calcium, iron, and potassium [4]. Several works have been done, reporting the levels of nutrients, including minerals in *Mo*. It found that the dried leaves had the following mineral contents: calcium (3.65%), phosphorus (0.3%), magnesium (0.5%), potassium (1.5%), sodium (0.164%), sulphur (0.63%), zinc (13.03 mg/kg), copper (8.25%), manganese (86.8 mg/kg), iron (490 mg/kg) and selenium (363 mg/kg) [4]. Research in Indonesia is still focused on nutritional value such as vitamins, proteins, carbohydrates, fiber, energy and limited to only some macro nutrients such as Ca, K and Fe, and not for micro nutrient content [5].

Trace elements are essential for living organisms, although they become toxic at high concentrations. Thus, both their deficiency and excess can be harmful to human health. The essential elements are elements required for good health and are required for normal body functioning that either cannot be synthesized by the body at all or cannot be synthesized in amounts adequate for good health and thus must be obtained from a food source. *Moringa* trees have been used to combat malnutrition especially among infants and breastfeeding woman in many developing countries, particularly in India, Pakistan, the Philippines, Hawaii and many parts of Africa [6]. It is particularly useful as a human food in tropical countries because the leaves appear towards the end of the dry season when few other sources of green leafy vegetables are not available [6].

In Indonesia, *Moringa* leaves are usually consumed as vegetables to improve nutrition in nursing mothers and children; used as traditional medicine such as lowering blood pressure and cholesterol, protecting liver against damage and diabetes. *Mo* plant can be grown in all regions in Indonesia. Cultivation is done in Kupang district NTT, Blora and Gunung Kidul. Several studies have been done, it was reported that nutritional content in the leaves of *Moringa* varies with a location where the plant is grown. There could be variations in the levels of macro and micro elements in the plant, depending on the soil properties of the geographical locations where it grows [7]. There are limited reports on the influence of variation in the geographical location of *Moringa* on the mineral composition and study of mineral content in the about *Moringa* here in Indonesia. Likewise, the study of mineral content in the micro- and trace order in *Moringa* leaves here is still rare, because it requires a sensitive analysis method and has low detection limits such as NAA and ICP. Considering these aspects, the objective of the present research was to investigate the mineral composed of *Moringa* leaves from Indonesian, as a preliminary study samples was taken from Blora to be compared with the results of studies conducted in several other countries. The data obtained can be used to estimate the intake of minerals obtained from the consumption of *Moringa* leaves. An analytical technique with sufficient sensitivity is required for accurate determination of elements in plant samples. Analytical techniques such as Neutron Activation Analysis (NAA) were used in this study.

MATERIAL AND METHODS

Sample Collection and Preparation

Samples were collected randomly from an area in Central Java. Samples were dried using freeze drier equipment at temperature -60 °C for 72 hours. The samples were powdered in an agate mortar and just passed through 100 mesh sieve and placed in a dark bottle. Special codes were given to each of the samples before storage.

Preparation of samples for neutron activation analysis

An amount of 50-100 mg of Mo leaves powder sample and reference material was weight accurately and packed in a polyethylene vial. Three replicates of each sample were prepared. Standard reference material of peach leaves 1547 from National Institute of Standards and Technology (NIST) and flux monitor of Al-0,1% Au from IRRM (*Institute for Reference Material and Measurement*) were prepared and irradiated together with the samples. The reference materials were used as a comparator standard for gamma spectrum evaluation using the relative method of standardization for neutron activation analysis (NAA) and to check for the accuracy of the analytical method used for k_0 -INAA.

Irradiation and Counting

Three samples together with standard packed in polyethylene container were irradiated for 2 minutes. Irradiation was carried out in Rabbit system of G.A. Sywabessy Multi-Purpose Reactor at Serpong, at a neutron flux of $\sim 10^{13} \text{ n cm}^{-2}\text{s}^{-1}$. After a period of time of 5 minutes cooling, the irradiated samples and standard were measured by high purity germanium detector from Canberra.

Samples were packed in polyethylene vial and aluminum foil for medium and long irradiation. Six samples of moringa leaf together with standard and flux monitor Al-0,1% Au from IRRM packed in an aluminum container were irradiated for a period of 30 minutes (medium irradiation) and 4 hours (long irradiation). After a period of time of 3 days (for medium irradiation) and 3 weeks cooling (for long irradiation), the irradiated samples, standard and flux monitor were measured by high purity germanium detector with 1.8 keV of resolution for 1332 keV of ^{60}Co and 25% relative efficiency from Canberra. The spectra were evaluated with the program Genie 2000 software version 3.2 [8]. Quantitative analysis was carried out by comparative- INAA for short half live radionuclide and k_0 -INAA for medium and long half live radionuclide.

RESULT AND DISCUSSION

Elemental data were validated by simultaneously analyzing reference material NIST 1547 peach leaves. A comparison of measured value and the NIST 1547 concentration of peach leaves are given in Table 1. Most of the measured value was less than $\pm 10\%$ different than the NIST concentration values, and those that were more than 10% different (Sc) were at very low concentrations that were near the detection limit. Some elements determined in the reference material are not certified (Rb, Br, Ce, La, Sm, Eu and Sc). From this result, INAA is a reliable method for determination of elements in Moringa leaf.

Table 1. Comparison of measured values of elemental concentration (mg/g) with certified values in peach leaves (NIST 1547)

Element	Measured value (mg/kg)	Certified value (mg/kg)	Difference (%)	Recovery (%)
K	23550 $\pm$ 1153	24300 $\pm$ 291	-3.09	96.91
Ca	15270 $\pm$ 1847	15600 $\pm$ 202	-2.12	97.88
Fe	213 $\pm$ 18	218 $\pm$ 14	-2.20	97.80
Ba	111 $\pm$ 11	124 $\pm$ 4	-10.08	89.91
Rb	20.40 $\pm$ 1.33	19.70	3.55	100.04
Zn	18.03 $\pm$ 1.64	17.90 $\pm$ 0.39	0.73	100.01
Br	11.38 $\pm$ 0.42	11.00	3.45	100.03
Ce	10.63 $\pm$ 0.62	10.00	6.30	100.06
La	9.04 $\pm$ 0.26	9.00	0.47	100.00
Sm	1.08 $\pm$ 0.04	1.00	8.20	100.08
Eu	0.19 $\pm$ 0.02	0.17	9.53	100.12
Sc	0.045 $\pm$ 0.001	0.04	12.50	100.13

The results analysis of samples by NAA confirming the presence of ten essential elements (Ca, Cl, Co, Cr, Fe, K, Mg, Mn, Na, Se, Br and Zn), two potentially toxic elements (Al and Sb) and eight other elements (Ba, Ce, Cs, La, Eu, Rb, Sc, and Sr) at a wide range of concentrations are presented in the Table 2. However, recent reports show that Br is an essential element [9]. Major elements, also known as macro minerals, are those elements which are needed in the body in quantities more than 100 mg per day. The macro minerals that are needed by the body to function include calcium, copper, phosphorus, potassium, sodium, and chloride. Minor minerals (trace elements) are needed in the body in less than 100 mg per day. Chromium, magnesium, iodine, iron, fluorine, manganese, selenium and zinc are examples of minor minerals. The macromineral found in samples was potassium 3345, calcium 3039, sodium 16.55 and chloride 253 mg/100g dry leaf Mo, respectively. The micro mineral in samples was chromium 0.476; magnesium 658; iron 14.72; manganese 10.21; selenium 0.008; and zinc 3.57 mg/100 g dry leaf. This result indicates that *Mo* leaves are a promising source of essential minerals [10]. The toxic elements, the concentration in the order as Al > Br > Sb, and the concentration of the other elements in order as Sr > Rb > Ba > Cs > La > Ce > Sm > Eu > Sc.

Table 2. Range and Mean Concentration of elements in the *Moringa oleifera* leaves

Element	Range Concentration (mg/100g)	Mean Concentration $\pm$ SD (mg/100g)
Ca	2787-3307	3039 $\pm$ 230
K	2672-4892	3346 $\pm$ 825
Cl	188-332	2531 $\pm$ 217
Mg	553-1566	658.77 $\pm$ 42.86
Sr	16.25-18.19	16.98 $\pm$ 1.06
Na	14.78-17.76	16.55 $\pm$ 1.26
Fe	14.11-16.57	14.72 $\pm$ 1.06
Rb	12.18-14.18	12.98 $\pm$ 0.24
Al	10.51-47.93	15.04 $\pm$ 1.77
Mn	8.62-11.72	10.21 $\pm$ 1.36
Ba	5.06-7.46	6.51 $\pm$ 0.92
Zn	3.32-3.88	3.57 $\pm$ 0.11
Br	0.44-0.50	0.48 $\pm$ 0.01
Cr	0.31-0.60	0.48 $\pm$ 0.02
Cs	0.15-0.17	0.16 $\pm$ 0.01
La	0.06-0.09	0.074 $\pm$ 0.002
Ce	0.046-0.080	0.063 $\pm$ 0.012
Co	0.016-0.018	0.017 $\pm$ 0.001
Sm	0.009-0.012	0.011 $\pm$ 0.001
Eu	0.002-0.004	0.003 $\pm$ 0.001
Se	0.002-0.022	0.008 $\pm$ 0.001
Sb	0.0013-0.010	0.010 $\pm$ 0.001
Sc	0.0003-0.0004	0.0003 $\pm$ 0.0001

The concentration of calcium (Ca) in the moringa oleifera sample is 3039 mg/100 g. Calcium is essential for a wide variety of functions in the body. It is present in the bones and teeth, and a small percentage is found in the blood and soft tissues, e.g. in the heart and kidneys, where it is responsible for nerve impulses and muscle contractions [11]. *Moringa oleifera* leaves are a very good source of potassium (K) 3346 mg/100g. K is found in blood plasma cell, muscles, and nerves. It regulates acid-base balance in the cell. Potassium reduces blood pressure [12]. Potassium works with sodium to maintain the water balance in the body and lowering the blood pressure. Sodium (Na) content in moringa is 16.55 mg/100g. Sodium is an important source of electrolytes within the body. Magnesium (Mg) level in *Moringa* leaves was 658.77 mg/100 g. Magnesium works with calcium to help to transmit nerve impulses in the brain. Mg is required in the plasma and extracellular fluid, wherein helping in maintaining osmotic equilibrium. Magnesium has a calming effect and works on the nervous system of those peoples, suffering from depression [13]. Zinc 3.57 mg/100 g can support the immune system and used for normal growth and development during pregnancy. Iron (Fe) 14.72 mg/100 g is the very important element as a nucleus

of hemoglobin that forms red blood cells in the body. Chromium level in moringa oleifera 0.48 mg/100 g. Chromium (Cr) is known to regulate carbohydrate, nucleic acid, and lipoprotein metabolism and it also potentiates insulin action. Deficiency of chromium decreases the efficiency of insulin and increases sugar and cholesterol in the blood. Chromium deficiency can cause an insulin resistance, impair in glucose tolerance and may be a risk factor for atherosclerotic disease [14]. Manganese (Mn) 10.21 mg/100 g is very useful for activation of some enzymes that prevent tissue damage and used for digestion and utilization of foods. Cobalt (Co) in moringa oleifera 0.017 mg/100 g is beneficial for humans because it is a part of vitamin B12, which is essential for human health. These obtained results agreed with that research before, whose reported that the moringa leaves are very promising source for essential elements [10,15]. The leaves of Mo are rich of nutrients, and is very important for its medicinal value. The Indonesian community is used Mo for supplements and traditional medicine for some diseases treatment.

The presence of several non-essential elements to the human body in Moringa Oleifera leaves, such as Ba, Ce, La, Sb, Sm, Sr, Eu, Rb and Al can be attributed from the soil, water or the atmosphere contamination. The concentrations of metals like Al and Sb involved in the biological cycle might cause serious damage to humans and other living organisms [16]. Aluminum is generally not regarded as an essential element and benefits of this mineral are not well documented. The result in this study compared with studies from other area are shown in Table 3.

This study confirmed that there are variations in macro and trace minerals in moringa oleifera leaves from different locations. This might be attributed to the variable uptake of minerals by the plant materials and variable agro-ecologies of the different regions [7,17]. Also, the variation in the nutritional values will differ for a wide range of reasons, such as cultivated regions, growing conditions, nature of the soil, seasonal changes, genetically different cultivars, storage conditions or due to the period of analysis [17]. From Table 3, it can be seen that the mineral content in the Mo leaf that grows in Central Java is similar in composition to those grown in India, Thailand, and Pakistan, but unlike in Namibia, Egypt, and Agra- India.

Table 3 Mean Concentration in this study compared with studies from other countries

Elements (mg/100g)	This study	Namibia [18]	Pakistan [17]	Agra- India [12]	Thailand [17]	India [20]	Nigeria [3]	Ghana [14]	Egypt [10]
Calcium	3039	242	2098	229	1904	2078	2600	3539	486
Magnesium	658	397	406	9.57	-	403	64.3	6099	25.64
Potassium	3345	639	1922	196	1795	1498	821	2522	33.63
Sodium	16.55	19.05	ND	-	-	72.50	298	3342	289
Iron	14.72	8.47	28.3	20.48	25.97	27.76	16.9	7.38	9.45
zinc	3.57	5.91	5.4	0.99	-	5.43	1.53	0.39	1.63
Cobalt	0.017	0.34	-	0.016	-	-	-	-	-
Mangan	10.21	1.77	-	7.48	-	32.45	6.99	0.012	5.21
Chromium	0.48	-	-	0.019	-	-	-	-	-
Arsenic	-	-	-	0.23	-	-	-	-	-
Aluminium	15.04	2.84	-	-	-	-	-	-	-
Barium	6.51	0.21	-	-	-	-	-	-	-

CONCLUSION

The research concluded that *Moringa oleifera* from central java is a nutrient-rich plant, especially in its leaves. It contains essential elements of Ca, Cl, Co, Cr, Fe, K, Mg, Mn, Na, Br, Se and Zn, make it a potential leaf source food that is suitable to combat malnutrition. It contains also toxic elements of Al and Sb and other elements are Ba, Ce, Cs, La, Eu, Rb, Sc and Sr. The mineral composition of Moringa oleifera from Central Java (Indonesia) significantly differs from others country. Determination of macro and micro mineral in the Moringa oleifera growing here should be done, to obtain more nutritional data especially for essential elements.

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