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Research article

Mineral Composition and Distribution Analysis of *Moringa oleifera* Lam. Seeds

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Abstract

Moringa oleifera seeds, in addition to serving as a means of propagation, are edible and contribute to human nutrition. Understanding their mineral composition and distribution is crucial due to the essential role of minerals in plant physiology and human health. This study employed micro-X-ray fluorescence (μ-XRF) techniques to analyze the composition and distribution of minerals in three treatments of *M. oleifera* seeds: the intact seeds (T1), the seeds without wings (T2), and the dehulled seeds (T3). The results identified eight minerals in the seeds: potassium (K), phosphorus (P), calcium (Ca), magnesium (Mg), sulfur (S), iron (Fe), silicon (Si), and aluminum (Al). There were significant differences in the content of Mg, S, K, Al, and Fe between treatments, while P, Ca, and Si did not show significant differences. Potassium was predominant in seeds with a seed coat, whereas sulfur was the most abundant in the seed kernel. Mineral distribution maps generated by μ-XRF analysis confirmed the localized concentration of specific minerals in specific seed parts. The variation in mineral composition among different seed components is associated with the functional roles of those parts. This finding also highlights the potential for selective use of *M. olifeira* seed parts based on their mineral content.

Keywords: μ-XRF analysis; mineral mapping; plant physiology; seed treatments; seed parts

1. Introduction

Moringa oleifera is a multipurpose yet underutilized plant that belongs to the Moringaceae family (Devkota & Bhusal, 2020; Tafesse et al., 2020). The tree originated in the Himalayas and has been introduced to various parts of the world (Devkota & Bhusal, 2020; Jattan et al., 2021). The spread of *M. oleifera* from its native range was related to its enormous benefits. All parts of *M. oleifera* are beneficial and can be used for various purposes, ranging from human food, animal fodder, and medicine (Devkota & Bhusal, 2020; Tafesse et al., 2020; Jattan et al., 2021). *Moringa oleifera's* high value is also due to its climate resilience (Horn et al., 2022), which will lead to an increasing distribution area under climate change scenarios (Bania et al., 2023).

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The seed is an essential part of *M. oleifera* as it serves as the plant's main generative propagation method (Jattan et al., 2021). However, the importance of *M. oleifera* seeds goes beyond their function for propagation; the seeds are also edible (Devkota & Bhusal, 2020; Tafesse et al., 2020; Jattan et al., 2021). This condition indicates that the seeds are of direct benefit to humans. *Moringa oleifera* seed's edibility also warrants a thorough study of the seed composition, including its mineral composition and distribution.

Minerals are essential for various plant physiological activities (Cervera-Mata et al., 2022), making them an essential constituent of plant organs, including the seeds. Minerals are distributed in specific locations of the seed (Cotrim et al., 2019; Montanha et al., 2022) and get redistributed during germination and seedling growth (Paul et al., 2020), a condition that highlights the importance of minerals in a plant's early life stage. However, the importance of seed minerals is more profound in edible seeds such as *M. oleifera* as they might support a healthy human diet (Cervera-Mata et al., 2022; Sahu et al., 2022). Thus, understanding *M. oleifera* seed mineral composition and distribution can directly and indirectly benefit humans.

To properly understand the *M. oleifera* seed minerals composition and distribution, a reliable mineral analysis technique is needed. X-ray fluorescence (XRF) stands as a possibility, as it is a widely used technique in mineral composition and distribution analysis (Ashe et al., 2025). XRF has already been used to study the mineral composition of various plant species and organs, including seeds (Feng et al., 2021; Montanha et al., 2022; Wibisono & Nurcholis, 2023). XRF has been used in the study of various parts of *M. oleifera,* including the leaves (Shaltout & Hassan, 2021), seed husks (Jami et al., 2019) and seed cake residues (Zaid & Ghazali, 2019). These reports highlight XRF's potential to analyze *M. oleifera* seed mineral properties.

Despite being widely used in mineral composition analysis of various plant species and organs, including *M. oleifera* seed, the application of XRF in the assessment of *M. oleifera* seed mineral distribution is still absent because research interest has been focused on the use of ground seeds or seed parts (Jami et al., 2019; Zaid & Ghazali, 2019). The use of grounded material prevents XRF from mapping mineral distribution. Previous studies (Jami et al., 2019; Zaid & Ghazali, 2019) did not compare the mineral composition of different *M. oleifera* seed parts, which has already been performed in soybean seeds (Wibisono & Nurcholis, 2023).

The mentioned condition presents knowledge gaps regarding *M. oleifera* seed mineral distribution and the comparison of the mineral composition of different parts of *M. oleifera* seeds. Thus, the present study aims to fill those gaps by mapping *M. oleifera* seed mineral distribution and its effect on the mineral percentage of different seed parts using the micro-X-ray fluorescence (μ-XRF) method. This research is done to supplement previous studies on *M. oleifera* seed mineral composition and to provide the first data on its localization.

2. Materials and Methods

This study uses *M. oleifera* seeds previously collected by a seed collection team from Bunutan Village, Abang District, Karangasem Regency, Bali Province, Indonesia (Figure 1). The seed collection team reported that the seed collection was conducted on 19 November 2022. The seeds were harvested from three *M. oleifera* individual plants in two collection points within the village. The first collection point was S 08.34741° and E 115.67967° at 29 m above sea level (m asl) altitude, while the second collection point was S 08.34788° and E 115.68040° at 67 m asl altitude. These *M. oleifera* individuals were grown in the coastal settlement area (Figure 1).

Figure 1. Seed collection sites. a. Bali Island with yellow rectangles represents Abang District; b. Bunutan Village; c. *M. oleifera* seed collection points. (Base maps from Google Earth)

During the harvesting period, mature seed pods were collected directly from the trees. Abiotic conditions during the collection period were 73.7% air humidity, 29.6°C air temperature, 1 knot wind speed, and a light intensity of 3,760 lux. Meanwhile, the tree substrate abiotic measurement shows 4.8 soil pH, and soil humidity of more than 80%. The plant and seed ages in the harvesting period were, unfortunately, unknown.

The harvested seed pods were kept at room temperature during the remaining duration of the field trip. At the end of the field trip period, these pods were then transported to the Bali Botanic Garden laboratory for storage. In the laboratory, the seeds were removed manually from the seed pods to prepare them for the drying process. During this extraction process, fifteen randomly selected seeds were separated for μ-XRF analysis. The selected *M. oleifera* seeds were divided into three groups. The first group was the intact seeds (T1), the second group was the seeds without wings (T2), and the last group was the dehulled seeds (T3) (Figure 2). This process was conducted at the laboratory ambient temperature of 23.5°C±1°C and humidity of 77%±3%.

The selected *M. oleifera* seeds were then cut in half using a pruner and packed into sealed plastic clips according to their treatment group. This procedure was performed due to the technical need for the μ-XRF analysis for flat surfaces. The packed seeds were then transported to the Nuclear Mineral Technology Laboratory-BRIN in Jakarta, Indonesia, for analysis. The μ-XRF assays were conducted on 12 January 2023; thus, there was a 54-day gap between the seed harvest and the μ-XRF analysis process.

The μ-XRF analysis was conducted by Nuclear Mineral Technology Laboratory staff using Bruker M4 Tornado Plus. The analysis was conducted on two *M. oleifera* seeds from each seed group, following the laboratory standard procedure. During this study, we treated the seed group as the treatment, while the number of analyzed seeds was treated as the replication for statistical analysis.

This study implemented both descriptive and statistical data analysis. Descriptive analysis was implemented to describe the mineral distribution of *M. oleifera* seeds. Meanwhile, statistical data analysis was conducted to understand the difference in seed mineral composition between treatments. This statistical analysis used one-way ANOVA, followed by a Tukey test at 5% if the ANOVA analysis found a significant result, and was conducted using JASP 0.19.3.0 statistical software (JASP Team, 2024).

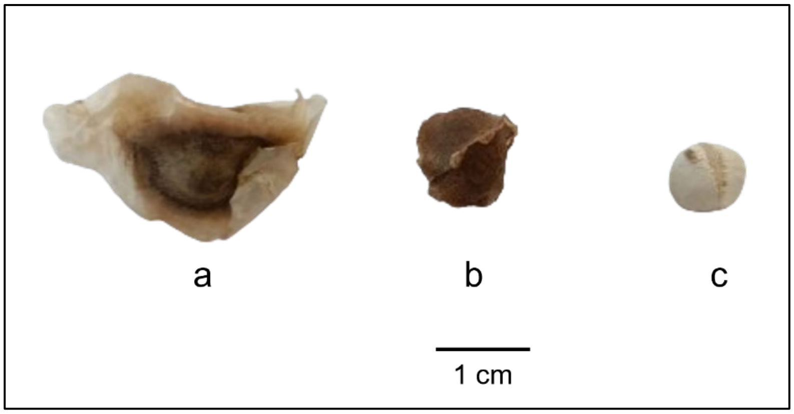

Figure 2. *Moringa oleifera* seeds for μ-XRF analysis. a. Intact seed; b. Seed without wings; c. Dehulled seed.

3. Results and Discussion

This study detected nine chemical elements in *M. oleifera* seeds. However, only eight minerals were detected, with the other chemical being oxygen. The minerals were the macrominerals such as potassium (K), phosphorus (P), calcium (Ca), magnesium (Mg), and sulfur (S), and microminerals such as iron (Fe), silicon (Si), and aluminum (Al). The minerals detected were also identified by previous studies using XRF to detect *M. oleifera* seed minerals composition and found calcium, potassium, sulfur, phosphorus, iron, manganese, and silicone (Jami et al., 2019; Zaid & Ghazali, 2019). However, the results in our studies were different from previous research; as we did not detect zinc, copper, molybdenum, and strontium while aluminum was detected. Zinc, copper, molybdenum, and strontium were present in *M. oleifera* seed, while aluminum was reported absent from previous studies (Jami et al., 2019; Zaid & Ghazali, 2019). This study also did not detect cobalt, which was detected in *M. oleifera* seed in previous studies, along with zinc, manganese, and copper using atomic absorption spectrophotometry (AAS) and inductively coupled plasma-optical emission spectroscopy (ICP-OES) (Stevens et al., 2021; Vinodkumar et al., 2025).

Despite the presence of dissimilarity to the previous studies, the relatively similar mineral composition in *M. oleifera* seed reported in this study is unsurprising as the studies were of the same species. However, the presence of macro and microminerals that were also detected in soybean seeds (Wibisono & Nurcholis, 2023) suggests that these minerals were contributors to plant seed minerals. Despite being needed in small amounts, micronutrients remain essential plant nutrients (Singh & Legese, 2022); thus, they are present along with macrominerals in the seed mineral composition.

This study also found that similar minerals were present in all the treatments. However, variety was found in the macromineral percentages. In T1 and T2, potassium was the mineral with the highest percentage, while in T3, sulfur was the highest percentage mineral, surpassing potassium as the second-highest percentage mineral. Despite sharing the same mineral as its highest percentage, the second-highest percentage of mineral in T1 and T2 differed with calcium and sulfur, respectively. Meanwhile, in T3, potassium was the second-highest percentage mineral. The treatments did not affect micromineral percentage, as the micromineral pattern was not different in all treatments, i.e., Si>Fe>Al.

Table 1 presents the mineral composition of *M. oleifera* seed with its mass concentration (normalization) [wt.%] value found in this study.

The results in this study showed potassium as the mineral of the highest percentage in *M. oleifera* seeds with seed coat (T1 and T2), which was in agreement with a previous study that mentioned the mineral as the highest percentage mineral in *M. oleifera* seeds (Vinodkumar et al., 2025). However, it is interesting to note that the previous study did not mention potassium as the most abundant mineral in *M. oleifera* seed husk (Jami et al., 2019). The result, which differed from the previous study, was also found in the dehulled seed, which recorded sulfur as the highest percentage mineral. While a previous study found calcium as the mineral with the highest percentage in the *M. oleifera* kernel (Zaid & Ghazali, 2019).

The difference in mineral composition between this study and previous studies might be attributed to several reasons. Although the effect of different sample origins on the composition and quantity of *M. oleifera* seed minerals cannot be overlooked, the sample condition during XRF analysis is another factor that should also be considered. XRF detects the elemental composition in the sample surface; thus, analysis on grounded seed parts, such as in previous studies (Jami et al., 2019; Zaid & Ghazali, 2019), can yield different results compared to analysis of unground seeds, such as those used in this study. However, the use of unground samples in XRF analysis was quite common, as it was used in the analysis of various plant species such as soybean and tef (Whisnant et al., 2024; Wibisono & Nurcholis, 2023). Using unground samples also enables XRF to determine the sample mineral localization.

This study's quantitative analysis shows that magnesium and sulfur percentages were significantly different between the treatments, while phosphorus, calcium, and silicon were not significantly different. Potassium and aluminum show similar significant patterns, with their percentages significantly different between T1 and T3 and not significantly different from T2. Meanwhile, the iron percentage in T3 was significantly different from that recorded in T1 and T2. This result suggests the presence of a specific mineral localization within *M. oleifera* seed, with some minerals more concentrated on certain seed parts than in others.

Table 1. Mass concentration (normalization) [wt.%] (mean±standard deviation) value in different *M. oleifera* seed treatments

Elements	**Treatments**		
	T1	**T2**	**T3**
Macromineral			
K	30.25±2.58 b	28.70±4.92 ab	16.32±0 a
P	3.46±0.58 a	3.77±0.15 a	4.81±0 a
Ca	15.52±6.77 a	7.76±0.52 a	2.56±0 a
Mg	5.43±0.21 c	2.36±0.07 b	1.40±0 a
S	8.98±1.99 a	15.95±1.89 b	24.87±0 c
Micromineral			
Fe	0.42±0.01 b	0.50±0.08 b	0.17±0 a
Si	0.75±0.12 a	0.60±0.08 a	0.47±0 a
Al	0.13±0.02 b	0.09±0.02 ab	0.03±0 a

Notes: Values followed by the same letter within the same row indicate no significant differences as determined by the Tukey test at a 5% significance level (p = 0.05); n = 2. T1=Intact Seeds; T2=Seeds Without Wings; T3=Dehulled Seeds; K=Potassium; P=Phosphorus; Ca=Calcium; Mg=Magnesium; S=Sulfur; Fe=Iron; Si=Silicon; Al=Aluminum.

The mineral localization maps acquired in this study were in agreement with the acquired quantitative data. Figure 3 shows the mineral distribution of *M. oleifera* seed minerals in different treatments. The localization images indicate that despite all minerals being present in all treatments, each treatment's primary mineral differed. The seed coat was dominated by potassium (purple); however, this mineral was only found in the periphery of the sulfur (cyan) dominated seed kernel. The mineral mapping also shows potassium as the primary mineral of the *M. olifeira* seed wing and coat, confirming the high percentage of potassium seeds with a seed coat. This condition also confirmed the insignificant difference in potassium values between T1 and T2 and its significant difference between T1 and T3. In contrast, the condition also confirmed the significant difference in the sulfur values of T1 and T3, highlighting the specific localization pattern of some minerals within *M. oleifera* seeds.

The high percentage of potassium and calcium in the *M. oleifera* seed coat agrees with the results of previous reports for tef, roselle, and soybean seed (Phewphong et al., 2023; Wibisono & Nurcholis, 2023; Whisnant et al., 2024). The result also agrees with previous findings for the *M. oleifera* seed husk (Jami et al., 2019). The high percentage of potassium and calcium is unsurprising, as they are among the major minerals in plants (Jing et al., 2024). The abundance of potassium and calcium in the *M. oleifera* seed coat is due to their role related to the seed coat's primary function, such as osmoregulation, water movement (Hasanuzzaman et al., 2018), and cell membrane integrity (Jing et al., 2024). On the other hand, the high percentage of sulfur in the dehulled seed is understandable as sulfur is important for seed metabolism and germination (Mondal et al., 2022). This finding indicates that mineral localization within *M. oleifera* seeds is correlated with the seed part function, indicating the importance of minerals in seed adaptation.

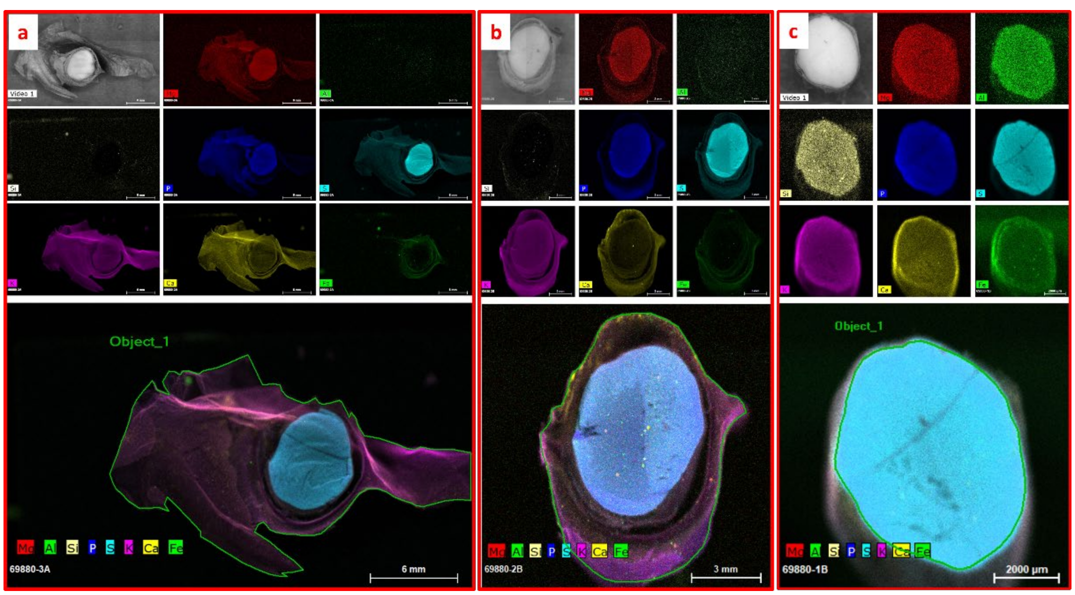

Figure 3. *Moringa oleifera* mineral mapping from µ-XRF analysis. a. Intact seed (T1); b. Seed without wings (T2); c. Dehulled seed (T3)

Even though mineral presence in plant organs, including the seed, is highly related to plant adaptation, humans can utilize this condition to their advantage. *Moringa oleifera* seeds are edible and can be utilized as a nutritious snack due to their high nutritional content (Gautier et al., 2022). This nutritional benefit might also be applied to the mineral-rich seed coat (Jami et al., 2019). The seed coat's high potassium concentration also opens up possibilities for the use of this part for seed priming purposes, as potassium is widely used in the seed priming procedure (Ali et al., 2020; Mebratu, 2022). This condition is also viable, as *M. oleifera* leaves are regarded as an effective seed priming agent (Soares et al., 2021). Furthermore, *M. oleifera* seed powder is considered a coagulant for water treatment (Omotesho et al., 2013). The seed aluminum content might support its use as an alternative synthetic aluminum coagulant (Krupińska, 2020).

Despite the enormous benefits offered by *M. oleifera* seed minerals, consideration needs to be given to the presence of hazardous minerals. Although absent in our study, toxic heavy metals, including lead (Pb), cadmium (Cd), and chromium (Cr), in the permissible range for human consumption were reported in a previous study (Sodamade et al., 2017). This further highlights the benefits of understanding the seed mineral composition of *M. oleifera* to enhance its utilization and safe consumption.

4. Conclusions

This study detected five macrominerals (K, P, Ca, Mg, and S) and three microminerals (Fe, Si, and Al) from *M. oleifera* seeds. The Mg, S, K, Al, and Fe percentages differed significantly between treatments, while P, Ca, and Si did not show significant differences. The quantitative and descriptive analyses of the study data indicated that different proportions of minerals were distributed in different parts of the *M. oleifera* seed, a condition that was related to the roles of minerals in supporting the seed part's function. This study fulfilled its aim of providing data regarding mineral composition and distribution in *M. oleifera* seed, enhancing our understanding of seed mineral function in plant species while also opening possibilities to improve the use of *M. oleifera* seed and seed parts concerning their mineral composition. However, toxic chemical screening and the use of *M. oleifera* from unpolluted planting sites should be considered, as toxic chemicals are reported in *M. oleifera* seed in previous studies, despite not being present in this study.

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6. Authors' Contributions

Farid Kuswantoro: Conceptualization, Methodology, Conducting the Research, Visualization, Validation, Writing – review and editing. Ayyu Rahayu: Statistical analysis, Writing – review and editing, Manuscript formatting.

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7. Conflicts of Interest

The authors declare that no conflict of interest is present in this paper.

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