# Extraction and Characterization of *Moringa oleifera* Leaves as a Feed Additive Candidate in Super Hybrid Duck Feed

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### Abstrak

Daun kelor (*Moringa oleifera*) atau dapat disebut dengan tanaman pagar merupakan salah satu tanaman yang banyak manfaatnya. Salah satunya yaitu dimanfaatkan sebagai *feed additive* pada hewan ternak yang bertujuan untuk mengoptimalkan pertumbuhan dan meningkatkan nilai gizi hewan ternak. Penelitian ini terfokus pada analisis zat yang terkandung dalam daun *Moringa oleifera*. Pertama daun *Moringa oleifera* mengandung kalsium tertinggi dengan kadar 68,4% dan 71,3%. Kedua, kaya akan kalium dengan kadar 18,3% dan 21,7% yang berguna untuk mengatasi fosfor dan sulfur pada hewan ternak. Ketiga, terdapat kandungan *Molybdenum* dengan kadar 7,8% yang berfungsi sebagai penawar racun. Keempat, memiliki kandungan protein sebesar 23,95% dalam pengukuran simplo dan 24,09% dalam pengukuran diplo dari hasil pengukuran protein dengan menggunakan metode Ketjel dan berfungsi meningkatkan kadar protein pada ternak.

### Keywords : Kelor, Daun, Pakan Additif

### Abstract

Moringa oleifera leaves usually called a hedge plant have many benefits. One of them is functioning as a feed additive in livestock aiming to optimize growth and increase the nutritional value of livestock. This study focused on analyzing the substances contained in Moringa oleifera leaves. First, Moringa oleifera leaves contain the highest calcium with levels of 68.4% and 71.3%. Second, they are rich in potassium with levels of 18.3% and 21.7%, as well as useful for dealing with phosphorus and sulfur in livestock. Third, there is a Molybdenum content with a level of 7.8% which functions as an antidote. Fourth, Moringa oleifera has a protein content of 23.95% in simplo measurements and 24.09% in diplo measurements from the results of protein measurements using the Ketjel method. Fifth, Moringa oleifera leaves potentially increase protein levels in livestock.

Keywords : Moringa oleifera, Leaves, Feed Additive

### INTRODUCTION

Based on data from the directorate general of livestock and animal health in 2021, the distribution of livestock in Indonesia was concentrated on Java Island. More specifically, cattle and buffalo, sheep and goats, as well as poultry are the largest populations in East Java. Based on data in 2020, these types of livestock increased by 4.81%, 2.13%, and 5.2%, respectively (Aslila Ramadhani D and Ledi Ermansyah 2021). However, the livestock population is unable to meet the needs of Indonesia's population continuing to grow every year. This is evidenced by the decrease in the number of cattle sent outside the East Java region. This condition implies that livestock farming should be improved. On the other hand, the quality of the beef produced is another essential factor, considering that animal protein is significant for consumption. In line with this, the quality of beef is influenced by its nutritional value. Therefore,

the production of high-quality meat meeting the biological needs of livestock with low feed costs is captivating to study.

Moringa oleifera often called the "Miracle tree" is a universal plant in which leaves. flowers and roots are useable for food and medicine. (Yagi et al. 2013). The availability of Moringa oleifera in Indonesia is abundant, yet its utilization is limited to hedges (shrubs) and it lives on river banks. (Manggara and Shofi 2018). Based on research studies in the last three years, many researchers concern about Moringa oleifera due to several constituent ingredients of the Moringa oleifera plant. (Arifin 2008; Magat, Raquepo, and Pabustan 2009; Fitriani, Walanda, and Rahman 2012). Moringa oleifera leaves contain 42% oil with unsaturated fatty acids of 75.8 - 82.9% with oleic acid as the main component of 71.2 - 79.5% (Rochmawati 2015). Meanwhile, Andrew et al. reported that Moringa oleifera seed is the second most substantial source of protein and nutrients after lipids (Hamzah and Yusuf 2019). The protein content in *Moringa oleifera* has been reported to vary between 18.6% and 37.2% (Bakri and Jayamani 2016), (Nandiyanto, Oktiani, and Ragadhita 2019). Then, Saa et al., 2010 also revealed that Moringa oleifera has an essential amino acid content of 30-40% (Taslim et al. 2019). Furthermore. Moringa oleifera seeds contain vitamins, such as provitamin, A. Bcomplex, and C, and essential minerals as macro elements, such as iron, copper, manganese, and zinc ("Karakteristik Tepung Daun Kelor Dengan Metode Pengeringan Sinar Matahari | Kurniawati | Prosiding Seminar Nasional Unimus" n.d.; Hariyanto et al. 2022). Due to the high protein content in Moringa oleifera, it is an ideal choice as a supplement in animal feed to obtain quality animal feed.

The method commonly used to elevate the nutrient content of *Moringa oleifera* leaves is fermentation treatment (Wang et al. 2019) and germination. [15]. Previous research conducted by Coello et al. demonstrated that germination for approximately 85.5 hours can enhance the highest protein and TPC content as well as later, maximize the antioxidant activity of *Moringa oleifera*. (Coello et al. 2018). However, this method requires quite a long time and its use is only on the seeds. Therefore, studies related to improving the nutrition of *Moringa oleifera* through methods that are simpler and relatively short in time are necessary to develop. In this study, increasing nutrition through testing of amino acids as a feed additive was carried out using a simple drying method on the leaves. *Moringa oleifera* plants are largely spread throughout Indonesia and the utilized to enhance the effectiveness of *Moringa oleifera*, especially in the leaves. The increased amino acid content in *Moringa oleifera* leaves is capable of not only being a Feed Additive candidate in Hybrid Super Duck Feed but also upgrading the economic value of *Moringa oleifera* leaves.

### METHOD

The picked *Moringa oleifera* leaves were then washed with clean water. After that, the *Moringa oleifera* leaves were dried at 30 - 35 °C and pounded with a simple machine. The obtained *Moringa oleifera* leaf powder was sifted using a 200 mesh sieve. Subsequently, the protein was tested using the Ketjel method by weighing one gram of *Moringa oleifera* leaf sample which had been sifted and mixed with selenium and concentrated H<sub>2</sub>SO<sub>4</sub>. Then, the destruction was carried out at 420 °C for one hour. The destruction results were then added with 50 mL and 40% NaOH. After that, 25 mL of 4% H3BO3 was placed as a reservoir in the distillation apparatus. The distillation process was carried out for four minutes. Next, the distillate was titrated with 0.2 NHCl solution until the color changed from green to red. The percentage of protein content (%) was calculated using equation 1 below:

Protein Levels (%) = 
$$\frac{(V_p - V_a) \times N \times 1.4007 \times Fk}{\text{Sample Weight (g)}}$$
 (1)

Notes:

 $V_p$  = Volume of HCl 0.2N for sample titration (mL) V<sub>b</sub> = Volume of HCl 0.2N for blank titration (mL) N = Normality of HCI 0.2N solution

Fk = Protein conversion facto

Furthermore, to determine the levels of amino acids of *Moringa oleifera* leaves, the amino acids were tested through UPLC.

# **RESULT AND DISCUSSION**

The *Moringa oleifera* leaf samples were extracted into powder. Therefore, the samples were easily characterized and dried to remove the water content without destroying the content in the *Moringa oleifera* leaves. During testing using XRF, there were two treatments given, namely by drying *Moringa oleifera* leaves at room temperature and combining them at 500 °C to decompose the organic compounds which can be indicated by the formation of ash. Then, the ash resulting from the furnace was indicated to be the result of minerals filling the cavity of the *Moringa oleifera* leaf cells. The minerals are in the form of free minerals and inorganic compounds (Sukirno and Murniasih 2009; Yagi et al. 2013). The addition of nitric acid to *Moringa oleifera* leaves heated at 500 °C aims to remove organic compounds and impurities to purely obtain inorganic compounds (minerals) (Manggara and Shofi 2018).

	Conc Unit (%)		
Compound	Room Temperature	Furnace 500 °C	
Р	1.1	1.9	
S	1.6	3.25	
K	18.3	21.7	
Ca	68.4	71.3	
Mn	0.37	0.24	
Fe	1.63	1.22	
Ni	0.05	0.10	
Cu	0.25	0.04	
Мо	7.8	0.1	
Yb	0.4	0.2	

Table 3. Result of XRF Test on Moringa oleifera Leaf Sample

The results of XRF characterization using showed the percentage (%) of the elemental content present in Moringa oleifera leaves as presented in Table 3. Based on the data in Table 3, there are ten types of macro and micro minerals composed in Moringa oleifera leaf powder. Macro minerals include Ca, P, K, and S. Macro minerals are specifically needed by livestock in larger quantities when compared to micro minerals. Meanwhile, microminerals are needed in small quantities yet play role in livestock's survival (Arifin 2008). The microminerals include Fe, Ni, Cu, Mo, Mn, and Yb. After that, both Moringa oleifera heated at room temperature and roasted at 500 °C had the highest calcium content with levels of 68.4% and 71.3%. Calcium is a macro mineral functioning to support bone growth, blood clotting process, heart muscle activator, enzyme activator, and protect the body from the absorption of radioactive substances (Magat, Raquepo, and Pabustan 2009; Fitriani, Walanda, and Rahman 2012). Furthermore, potassium is the second highest in Moringa oleifera leaves with levels of 18.3% and 21.7%. Potassium plays a role in regulating muscle contractions, catalysts, and controlling heartbeat (Fitriani, Walanda, and Rahman 2012; Rochmawati 2015). In addition, the next two minerals are phosphorus and sulfur. Both minerals present in Moringa oleifera leaves can be used as an alternative to overcome the deficiency of phosphorus and sulfur in livestock's bodies even though the levels are low.

Moreover, the iron mineral is the second highest micromineral found in the content of *Moringa oleifera* leaves with room temperature treatment. Meanwhile, the iron mineral decreased in *Moringa oleifera* leaves heated at 500 °C. Iron mineral substantially functions to form the livestock's immune system (Hamzah and Yusuf 2019). The relatively high iron content when compared to other micro-minerals has the potential to meet the livestock's iron needs.

Molybdenum was found in *Moringa oleifera* leaves with levels of 7.8% at room temperature and 0.1% at 500 °C. Molybdenum acts as an antidote in the body. Other confirmed microminerals are Mn, Ni, Cu, and Yb with relatively small levels.

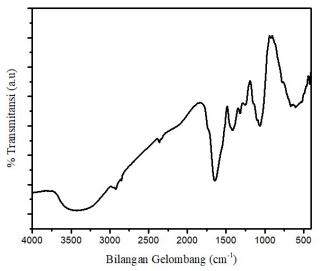


Figure 2. Functional Group of Moringa oleifera

Figure 2 shows the organic compound content of *Moringa oleifera* leaves. It has been confirmed from the IR spectrum that stretching, asymmetric vibrations, shape changes, fracturing, and blending with water occur in the functional groups of *Moringa oleifera* leaves. At wavenumbers of 3406 cm<sup>-1</sup>, 2924 cm<sup>-1</sup>, 1797 cm<sup>-1</sup> stretching occurs in the functional groups of O-H, C-H, and C=C, respectively (Bakri and Jayamani 2016; Nandiyanto, Oktiani, and Ragadhita 2019). Then, at a wavenumber of 1429 cm<sup>-1</sup>, there is the shape change of the C-H asymmetric vibration. At the wavenumbers of 1149 cm<sup>-1</sup> and 711 cm<sup>-1</sup> the C-H and CH2 vibration shape change as well (Nandiyanto, Oktiani, and Ragadhita 2019). Furthermore, the CH=CH vibration frame occurs at a wavenumber of 602 cm<sup>-1</sup>(Taslim et al. 2019). The presence of peaks at these wavenumbers indicates that *Moringa oleifera* leaves contain organic compounds with functional groups binding inorganic and organic ions.

Wavenumber (cm <sup>-1</sup> )	Functional Group				
3406	OH				
2924	CH	Stretching			
1798	C=C	-			
1429	СН	Asymmetry			
1149	CH	Vibration	shape		
711	CH <sub>2</sub>	change			
602	CH=CH	Vibration frame			

In this study, the protein was tested using the Ketjel method to certainly determine the total protein content of *Moringa oleifera* leaves. The protein measurements using the Ketjel method revealed protein levels of 23.95% and 24.09% for simplo and diplo measurements, respectively. In previous studies, the protein content of *Moringa oleifera* leaves heated at room temperature has also been reported. Using a different method (Bradford), namely UV-Vis spectroscopy, the total protein content obtained was 9% (Hariyanto et al. 2022). Based on these results, the Ketjel method is more effective in determining the protein content of a material. The results of measurements using the Ketjel method are in accordance with

research conducted by Kurniawati et al., 2018, reporting that the protein content of sun-dried *Moringa oleifera* leaves was 23.37% (Kurniawati, Fitriyya, and Wijayanti 2018).

No.	Doromotor	Unit	Type of	Result	
INO.	Parameter		Amino Acids	Simplo	Duplo
1	L-Serine	L-Serine mg/kg		11619.18	11606.78
2	L-Glutamic Acid	mg/kg		29069.04	29169.39
3	L-Phenylalanine	mg/kg		12316.79	12350.05
4	L-Isoleucine	mg/kg	g g Essential g g	8424.75	8474.77
5	L-Valin	mg/kg		10526.83	10590.42
6	L-Alanine	mg/kg mg/kg		10686.77	10758.60
7	L-Arginine			12236.53	12226.03
8	Glycine	mg/kg		10607.25	10622.49
9	L-Lysine	mg/kg		12740.01	12792.78
10	L-Aspartic Acid	mg/kg		23361.21	23414./7
					0
11	L-Leucine	mg/kg		17501.44	17568.21
12	L-Tyrosine	mg/kg		6801.03	6807.41
13	L-Proline	mg/kg	non essential	9313.59	9363.55
14	L-Threonine	mg/kg		11417.30	11457.98
15	L-Histidine	mg/kg		4314.48	4270.91

Table 5. Result of Test on Amino Acids of Moringa oleifera Leaf Sample

The amino acid test results are presented in Table 5. Based on Table 5, there are two types of amino acids contained in *Moringa oleifera* leaves, namely essential and non-essential amino acids. The essential amino acids contained in *Moringa oleifera* leaf nano powder include L-serine, L-glutamic acid, L-Phenylalanine, L-Isoleucine, L-Valine, L-Alanine, L-Arginine, Glycine, L-Lysine, and L- Aspartic acid. Meanwhile, the non-essential amino acids contained in *Moringa oleifera* include L-Leucine, L-Proline, L-Proline, L-Threonin, and L-Histidine. These results imply that the 15 kinds of amino acids contained in *Moringa oleifera* leaves can support increasing protein levels in animal feed.

### CONCLUTION

Based on research results, *Moringa oleifera* leaves contain the highest calcium with levels of 68.4% and 71.3%. They also contain potassium with levels of 18.3% and 21.7% which are useful for dealing with phosphorus and sulfur in livestock. There is a Molybdenum content with a level of 7.8% functioning as an antidote. In addition, *Moringa oleifera* leaves are rich in protein. The protein measurements using the Ketjel method obtained protein levels of 23.95% and 24.09% for simplo and diplo measurements, respectively. The protein can increase protein levels in livestock.

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