

# Addition of *Aspergillus Niger* Palm Waste (Animal Feed Alternative)

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## ABSTRACT

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The study aimed to determine the effect of adding the fungus *Aspergillus niger* to the chemical quality of fermented palm oil leaf and midrib waste as an alternative feed ingredient. The research design used a Completely Randomized Design (CRD) in one direction with four treatments and four replications. The treatment given to oil palm leaves and midribs was P0 = oil palm leaves and midribs + 20% EM4, P1 = oil palm leaves and midribs + 20% EM4 + 1% *Aspergillus niger*, P2 = oil palm leaves and midribs + 20% EM4 + 2% *Aspergillus niger*, P3 = oil palm leaf and midrib + 20% EM4 + 3% *Aspergillus niger*. This fermentation was carried out using the Least Significant Difference Test (SDT) method. The results showed that the fermentation of oil palm leaves and midribs could significantly increase the crude protein content in the P2 treatment (10.32%) and organic matter in the P1 treatment (29.09%), but had no significant effect on dry matter and crude fiber.

**Keywords:** *Aspergillus Niger*, Waste, Animal Feed.

## I. INTRODUCTION

The availability of forage sources is decreasing due to the use of land for housing and the cultivation of food crops that can be used directly for human needs. The utilization of agricultural waste as an alternative feed is a solution to overcome the shortage of ruminant feed. The by-products that are often considered as waste from plantations are oil palm leaves and midribs. Because in general the success of the livestock business must be supported by the availability of feed to meet basic needs, production and reproduction specially to

meet the needs of forage feed in the dry season by utilizing the by-products of plantation activities.

The by-products of plantation crops that have not been utilized optimally are the by-products of oil palm plantations [1],[2]. This plantation plant has the potential to be used as feed for ruminants in the form of oil palm leaves and midribs [3],[4]. This by-product contains dry matter, crude protein, and crude fiber whose nutritional value can be used as a basic ingredient for ruminant animal feed [5]. Oil palm leaves and midribs are one of the by-products of plantations resulting from oil palm pruning whose utilization has received little attention from farmers [6].

Oil palm leaves and midribs can be used as a source of forage reserves for ruminants [7]. Oil palm leaves and midribs can be used as substitute feed ingredients. The largest constituent components of palm fronds consist of cellulose, hemicellulose, lignin, and silica [8],[9],[10]. Oil palm fronds have been a by-product of plantations originating from oil palm pruning with each pruning consisting of 1-3 fronds per tree and having great potential to be utilized as feed [11],[12]. Each hectare of oil palm plantations has 148 trees with the number of midribs produced reaching 3,500-1,600 per year and producing oil palm leaves of 3.3 kg of fresh leaves, 35% dry matter, or 34,5 kg dry matter/ha/day.

Increasing the nutritional value and digestibility of palm frond waste is through the fermentation process [13],[14]. Fermentation is the process of chemical changes in organic substrates by the presence of biochemical catalysts, namely enzymes produced by certain types of microorganisms [15]. The process supports microbial growth during the fermentation process [1]. Changes during the fermentation process are largely influenced by the activity of microorganisms that cause changes in improving the quality of feed ingredients both from the nutritional aspect as well as digestibility and storage [16],[17],[18]. This increase can be done by utilizing biological feed technology, namely *Aspergillus niger* [2].

*Aspergillus niger* is a fungus from ascomycete that is filamentous, has hyphae, branched and insulated, light or colorless, and is found abundantly in nature [19],[20],[21]. Fungi are isolated from soil, plant debris, and indoor air [22]. *Aspergillus niger* grows optimally at a temperature of 35-37°C, with a minimum temperature of 6-7°C and a maximum temperature of 45-47°C with the growth process of this fungus is aerobic. *Aspergillus niger* has a white or yellow base color with a thick layer of conidiospores, dark brown [23],[17]. Metabolism *Aspergillus niger* can produce citric acid and can grow quickly so it is widely used

commercially in the production of citric acid, gluconic acid, and the manufacture of enzymes such as amylase, pectinase, amyloglucosidase, and cellulase [24],[25]. Bio activators in the *Aspergillus niger* mushroom fermentation process can help the bioconversion process of organic and inorganic compounds into a protein so that the fermented substrate content increases and can break down cellulose, xylanase, and hemicellulose fibers in the crude fiber content of palm oil waste [26],[27],[28].

Southeast Sulawesi is an area that has oil palm plantations covering an area of 7,459 ha and Konawe Selatan Regency reaching 1,055 ha which has by-products in the form of oil palm leaves and midribs that can be used as an alternative feed for ruminants because they contain nutrients. The palm oil waste can be used as an alternative feed ingredient. However, the use of oil palm leaves and midribs as animal feed ingredients is still limited due to the high crude fiber content and low digestibility. So, it is necessary to apply fermentation technology in the processing of palm oil leaves and midribs which can help increase the digestibility of the feed ingredients. The background of this study provides a little description of the potential of palm oil waste as an alternative source of animal feed, but there are obstacles, namely crude fiber which is high enough so that an alternative to solve this problem is to use the fungus *Aspergillus niger* as a bio activator to determine the effect of the chemical quality of fermented oil palm leaves and midribs.

## II. METHODS AND MATERIAL

### 2.1 Time and Place

The research was carried out for two months from February 27 to April 2022, consisting of two stages, namely the first stage covering the fermentation of palm oil leaves and midribs, the second analyzing the nutritional content of research materials conducted at the Laboratory of Nutrition Science and Feed

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## 2.2 Research Material

The equipment used in this study were: chopper, glass jar, bucket, dipper, tarpaulin, scale, porcelain dish, plastic bag, measuring cup, petri dish, vacuum pump, desiccator, oven, furnace, Erlenmeyer, whattman filter paper no. 41, measuring flask, distillation flask, Kjeldahl flask, fume hood, electric heater, centrifuge, mohr pipette, drip pipette, a set of distillation apparatus, and analytical balance.

The materials used in this study were: oil palm leaf midrib that was  $\pm$  4 years old, EM4, *Aspergillus niger* mushroom, granulated sugar, aqua dest, phloroglucinol 1%, H<sub>2</sub>SO<sub>4</sub>, selenium, pepsin, 0.01 N HCL, NaOH, BCG-MR.

## 2.3 Research Procedure

The collection and preparation of oil palm leaf and midrib materials in this study by taking samples of oil palm leaves and midribs with an age of  $\pm$  4 years at random from each oil palm tree, counting the by-products of palm oil leaves and midribs with a size of 1-2 cm and drying the material. which has been chopped on a tarpaulin for 12 hours.

After taking and preparation, the next step was to activate the EM4 solution by mixing 25 ml of EM4 solution with 2500 ml of water, and 250 g of granulated sugar, then incubating closed for 24 hours. After 24 hours, the solution is mixed with oil palm leaves and midribs with a ratio of every 3000 grams of oil palm leaves and midribs will be sprayed with 600 ml of activated EM4 solution, a mixture of oil palm leaves and midribs and EM4 is then taken and mixed with *Aspergillus niger* fungus according to with the

treatment of 0%, 1%, 2% and 3% of the dry weight of the leaves and midrib of oil palm, these materials are then fermented for 21 days and after twenty-one days, then they are opened and taken as samples for each analysis treatment proximate.

## 2.4 Research Design

This study was arranged based on a Completely Randomized Design (CRD) with a unidirectional pattern with four treatments and each treatment was repeated four times. The treatment in this study is as follows:

P0 =Palm Oil Leaf and Midrib + 20% EM4.

P1 =Palm Oil Leaf and Midrib + 20% EM4 + 1% *Aspergillus niger*.

P2 =Palm Oil Leaf and Midrib + 20% EM4 + 2% *Aspergillus niger*.

P3 =Palm Oil Leaf and Midrib + 20% EM4 + 3% *Aspergillus niger*.

## 2.5 Observation Variable

The variables observed in this study were the content of dry matter (DM), organic matter (OM), crude fiber (CF), and crude protein (CP).

## 2.6 Data Analysis

The data obtained will be analyzed for variance (ANOVA), if there is a significant effect, it will be carried out with the smallest significant difference test (SDT)

## III.RESULTS AND DISCUSSION

The average dry matter (DM), organic matter (OM), crude fiber (CF), and crude protein (CP) content of fermented oil palm leaves and midribs using the fungus *Aspergillus niger* are presented in Table 1.

**Table 1.** Average dry matter content, organic matter, crude fiber, and crude protein of fermented oil palm leaves and midribs using *Aspergillus niger* fungus

Parameter	Treatment			
	P0	P1	P2	P3
Dry Matter (DM)	29.55±0.42	30.16±0.41	30.63±1.35	29.40±0.45
Organic Matter (OM)	23.12±3.20 <sup>a</sup>	29.09±2.47 <sup>b</sup>	27.51±1.77 <sup>ab</sup>	27.40±1.34 <sup>ab</sup>
Crude Fiber (CF)	24.59±1.18	24.03±1.27	22.79±1.55	24.74±0.78
Crude Protein (CP)	8.84±0.35 <sup>b</sup>	9.28±0.47 <sup>b</sup>	10.32±0.27 <sup>c</sup>	7.66±0.69 <sup>a</sup>

**Note:** <sup>ab</sup>Different superscripts on the same line show significantly different treatments ( $P < 0.05$ ) based on the follow-up test (BNT). P0: Control treatment (EM4 20%), P1: EM4 20% and *Aspergillus niger* 1%, P2: EM4 20% and *Aspergillus niger* 2%, P3: EM4 20% and *Aspergillus niger* 3%.

### 3.1 Dry Matter (DM)

The results of this study are thought to be due to the addition of additives that cause an increase in the water content during the fermentation process so that it affects the dry matter, the higher the water content during the fermentation process, the loss of dry matter content increases. The increase in additive levels is thought to stimulate fermentation activity, causing H<sub>2</sub>O production to also increase [29]. The increase in dry matter is influenced by the increase in water content from fermentation [30],[31].

One of the factors that influence the dry matter content is the smaller the size of the substrate, the less the water element used as a growing medium for the *Aspergillus niger* fungus. The increase in dry matter due to cellulose degradation by inoculum was influenced by the growth of the fungus *Aspergillus niger*. However, in this study using a substrate size of 1-2 cm in the process of fermenting palm oil leaves and midribs, is thought to be the cause of not increasing dry matter content. The use of fine particle size resulted in a higher dry matter content than the fermentation product and was caused by the effectiveness of dry matter degradation during the fermentation process which was better on the surface area of fine particles [32],[33]. The smaller substrate particle size will

increase the contact between fungal cells and their mycelium with the substrate so that it can increase cellulase enzyme activity.

### 3.2 Organic Matter (OM)

Organic matter is a dry matter that has been reduced by ash and is present in feed available in an insoluble form. The results of the analysis of variance showed that the fermented treatment of palm fronds and leaves using the *Aspergillus niger* fungus which was different for each treatment had a significant effect ( $P < 0.05$ ) on the organic matter content of the midrib and oil palm leaves. P1 treatment had a significant effect on P0 but was not significantly different from P2 and P3, so it can be said that P1 treatment with *Aspergillus niger* 1% could increase the organic matter content compared to P2 and P3 treatments. The high content of organic matter in P1 treatment was because there were not many elements in the substrate that were utilized by the fungus *Aspergillus niger* in P1 treatment so there were still a lot of elements that made up the organic matter of the substrate. Meanwhile, in the P3 treatment with *Aspergillus niger* 3%, the value of the organic matter content was lower than in the P1 and P2 treatments, presumably because the higher the microbial dose, the more microbes degraded the substrate.

This study illustrates that the higher the dose of *Aspergillus niger* fungus given, the lower the value of organic matter content [34], This is because the energy needed for the growth of the fungus is increasing so that the fungus utilizes the elements of organic matter [35]. The more mushrooms that grow and are produced by enzymes that help in breaking down food substances for fungal growth it will affect the organic matter content of the substrate [36],[37],[38].

### 3.3 Crude Fiber (CF)

Crude fiber is part of carbohydrates, mostly derived from plant cell walls, and contains cellulose, hemicellulose, and lignin. The crude fiber in the form of cellulose, hemicellulose, and pectin is associated with lignin in the cell walls of feed plants and serves to strengthen the structure of plant cells [39],[40]. The presence of these structures in plants makes them the main source of crude fiber needed for ruminants.

Table 1 shows that fermentation using the fungus *Aspergillus niger* had no significant effect ( $P > 0.05$ ), on the crude fiber content of fermented palm midribs and leaves. This shows that the fermentation treatment with *Aspergillus niger* fungus 1%, 2%, and 3% does not affect the crude fiber content of fermented palm oil leaves and midribs. *Aspergillus niger* mushroom aims to increase protein, especially for dry ingredients that are low in protein. The content of crude fiber has a negative relationship with digestibility, the lower the crude fiber, the higher the digestibility of the ratio [41]. Crude fiber has the greatest influence on the digestibility of cellulose and hemicellulose which are difficult to digest, especially if they contain lignin [42]. An increase in the level of crude fiber will cause a decrease in the level of digestibility [43], thus consuming more feed to meet their energy needs [44].

Another cause of crude fiber does not have a real effect because the dose used is still low which results in the enzymes produced by the fungus *Aspergillus niger* not being able to secrete the cellulase enzyme to degrade

the fiber of palm leaves and midribs. The decrease in fiber content in substrate fermentation is thought to be due to, among other things, the relatively low dose of *Aspergillus niger* used in fermentation and *Aspergillus niger* used is not suitable for the type of substrate or is not compatible so that the results are less than optimal. The use of different *Aspergillus niger* in the fermentation process of palm fronds and leaves is caused by the ability of fungi or molds from microbes that have not been able to secrete various enzymes, especially cellulase enzymes to degrade fiber in oil palm midribs.

### 3.4 Crude Protein (CP)

Crude protein is all substances containing nitrogen by an average of 10% (range 13-19%). The results of the analysis of variance showed that the fermentation treatment of oil palm leaves and midribs had a very significant effect ( $P < 0.01$ ) on the crude protein content of oil palm leaves and midribs. The treatments P0 and P1 were not different but P3 and P2 were different for all treatments. This shows that the P2 treatment can increase the crude protein content because during the fermentation process there is a chemical and physical process on the substrate material and due to the activity of the fungus *Aspergillus niger* which ferments the fiber in the substrate using the cellulase hydrolysis enzyme which is then used for growth so that the crude protein content is reduced. in the P2 treatment was higher than the other treatments. The use of *Aspergillus niger* mushroom with a concentration of 2% by weight of dry matter can increase the crude protein content [45]. Fermentation with *Aspergillus niger* has been shown to increase protein and small peptide levels and eliminate trypsin inhibitors.

The increase in crude protein content during the fermentation process is because each fungal cell is a protein. The more fertile the mushroom grows, the higher the protein content [46]. The fungal cell wall contains 6.3% protein, while the cell membrane in hyphal fungi contains 25-45% protein and 25-30%



carbohydrates [47],[21],[48],[37]. The growth of fungi uses carbon and nitrogen for the cell components of the fungal body. This process occurs because, during fermentation, the fungus *Aspergillus niger* uses nutrients (especially carbohydrates) for growth.

#### IV.CONCLUSION

The results of the study concluded that the fermentation of oil palm leaves and midribs using the *Aspergillus niger* fungus could increase the crude protein content in the P2 treatment (10.32%) and Organic Matter in the P1 treatment (29.09%), while the dry matter and crude fiber analysis did not have a significant effect true to all treatments.

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