Evaluation of Enzyme Activity and Nutrient Content of Fermented Coconut Meal as a Feed Using Indigenous Microorganisms

Hera Dwi Triani[1*], Gusri Yanti[1], Wulansih Dwi Astuti[2], Sri Suharti[3], and Yetti Marlida[4]

[1]Faculty of Sains and Sosial, Prima Nusantara University, Bukittinggi, Indonesia
[2]Research Center for Applied Zoology, National Research and Inovation Agency (BRIN), Bogor, Indonesia
[3]Faculty of Animal Science, IPB University, Bogor, Indonesia
[4]Faculty of Animal Husbandry, Andalas University, Padang, Indonesia
***Corresponding author's email: heratriani@gmail.com**

Abstract. Coconut pulp has potential as animal feed but is limited in poultry diets due to its high fiber content and low protein levels. Fermentation using indigenous microorganisms offers a practical approach to improving its nutritional value. This study aimed to evaluate the nutritional quality of coconut meal using indigenous microorganisms (IMOs). The initial phase of the study involved producing IMOs, followed by measuring pH, total colony, and cellulase and mannanase enzyme activities. IMOs were applied to ferment coconut meal using a completely randomized design with four treatments (0, 7, 14, and 21 days) and four replications. Observed parameters included cellulase and mannanase enzyme activities, pH, crude fiber, crude protein, crude fat, and nitrogen-free extract. The characteristics of IMOs were analyzed descriptively, while enzyme activities and the nutritional content of coconut meal were analyzed using ANOVA and Duncan's Multiple Range Test. The results showed that IMOs had a pH of 3.3, were dominated by lactic acid bacteria with a total colony of 5.4×10^{12} CFU/mL, had cellulase activity of 2.1 U/mL, and had mannanase activity of 7.73 U/mL. Cellulase activity at 7, 14, and 21 days was significantly higher ($P<0.01$), while mannanase activity was highest at 21 days ($P<0.01$), with the lowest pH observed at 14 and 21 days. The lowest crude fiber and crude fat content, as well as the highest crude protein content ($P<0.01$), were observed at 14 days of fermentation. Fourteen days of fermentation were identified as the optimal duration for enhancing the nutritional quality of coconut pulp.

Keywords: cellulase, coconut pulp, IMOs, mannanase, nutrient

Abstrak. Ampas kelapa memiliki potensi sebagai pakan ternak tetapi terbatas dalam pakan unggas karena kandungan seratnya yang tinggi dan kadar protein yang rendah. Fermentasi menggunakan mikroorganisme asli menawarkan pendekatan praktis untuk meningkatkan nilai gizinya. Penelitian ini bertujuan untuk mengevaluasi kualitas gizi bungkil kelapa menggunakan mikroorganisme asli (IMO). Tahap awal penelitian melibatkan produksi IMO, diikuti dengan pengukuran pH, total koloni, dan aktivitas enzim selulase dan mananase. IMO diaplikasikan untuk memfermentasi bungkil kelapa menggunakan rancangan acak lengkap dengan empat perlakuan (0, 7, 14, dan 21 hari) dan empat ulangan. Parameter yang diamati meliputi aktivitas enzim selulase dan mananase, pH, serat kasar, protein kasar, lemak kasar, dan ekstrak bebas nitrogen. Karakteristik IMO dianalisis secara deskriptif, sedangkan aktivitas enzim dan kandungan gizi bungkil kelapa dianalisis menggunakan ANOVA dan Uji Jarak Berganda Duncan. Hasil penelitian menunjukkan bahwa IMO memiliki pH 3,3, didominasi oleh bakteri asam laktat dengan total koloni $5,4 \times 10^{12}$ CFU/mL, memiliki aktivitas selulase 2,1 U/mL, dan memiliki aktivitas mananase 7,73 U/mL. Aktivitas selulase pada hari ke-7, ke-14, dan ke-21 secara signifikan lebih tinggi ($P<0,01$), sedangkan aktivitas mananase tertinggi pada hari ke-21 ($P<0,01$), dengan pH terendah teramati pada hari ke-14 dan ke-21. Kandungan serat kasar dan lemak kasar terendah, serta kandungan protein kasar tertinggi ($P<0,01$), teramati pada hari ke-14 fermentasi. Fermentasi selama empat belas hari diidentifikasi sebagai durasi optimal untuk meningkatkan kualitas nutrisi ampas kelapa.

Kata kunci: selulase, ampas kelapa, IMO, mananase, nutrisi

Introduction

Coconut pulp is one of the wastes that has the potential to be used as an unconventional feed ingredient. Indonesia is one of the largest coconut producers globally, with a plantation area of 3.3 million hectares and coconut production reaching 2,800,000 tons (BPS, 2024). Coconut pulp constitutes approximately 70% of coconut meat. However, its low nutritional quality limits its use as feed (Sundu et al., 2020). The crude fiber content of coconut pulp is as high as 28.72% (Hidayati, 2017), with crude fat at 38.24% and protein at only 5.86% (Laksono et al., 2023). The use of coconut pulp in poultry rations

is not optimal because of the presence of fibrous carbohydrates in the form of mannose-based polysaccharides known as mannan (Sundu et al., 2020), which inhibits feed digestibility in the poultry digestive tract. Various methods to manipulate nutrition, such as the addition of the amino acids lysine and methionine or physical modification, have been attempted; however, these approaches have proven less effective. The fermentation method by microbes is considered effective for hydrolyzing cellulose and mannan because microorganisms serve as the primary producers of hydrolytic enzymes due to their fast growth rate, ease of cultivation, high enzymatic activity, and stability (Hasanah et al., 2024). Effective fermentation depends on the microbial strain and enzyme activity for hydrolyzing the substrate. The study prior to fermentation using *Aspergillus niger* showed no significant reduction in the crude fiber content of coconut meal (Laksono et al., 2023).

The fiber fraction in coconut pulp predominantly consists of cellulose and mannan, which can be hydrolyzed by the enzymes cellulase and mannanase. Cellulose is hydrolyzed into glucose, while mannan is degraded into manno-oligosaccharides and smaller amounts of mannose, glucose, and galactose (Patel et al., 2024). Fermentation using inoculants with mannanase and cellulase enzyme activities offers a more effective method to improve the quality of coconut pulp. The use of indigenous microorganisms (IMOs) with mannanase and cellulase enzyme activities as inoculants in coconut pulp fermentation provides a cost-effective and practical solution.

IMOs are bioactivators that can enhance the quality of agricultural waste when used as feed. They contain various enzymes essential for feed fermentation (Yulianto et al., 2022). Indigenous microorganisms are derived from natural sources containing beneficial microbes such as *Rhizobium sp*., *Azospirillum sp*., *Azotobacter sp*., *Pseudomonas sp*., and *Bacillus sp*. (Khasanah et al., 2019). The enzyme content in IMOs depends on the microorganism present, as they are typically isolated from their substrates (Yunilas et al., 2021). IMOs are bioactivators that can enhance the quality of agricultural waste for use as feed. They contain various enzymes essential for feed fermentation (Yulianto et al., 2022). Indigenous microorganisms are derived from natural sources containing beneficial microbes such as *Rhizobium sp., Azospirillum sp., Azotobacter sp., Pseudomonas sp., and Bacillus sp.* (Khasanah et al., 2019). The enzyme content in IMOs depends on the microorganism present, as they are typically isolated from their substrates (Yunilas et al., 2021).

IMOs produced from coconut by-products, such as coconut water and meal, have the potential to stimulate the growth of microbes with mannanase and cellulase enzyme activities due to the high crude fiber and mannan content in coconut meal. Coconut pulp contains 61% galactomannan and 26% mannan (Bahri et al., 2018). The relatively high galactomannan and mannan content in coconut meal can be utilized to produce mannanase and cellulase enzymes (Hatta et al., 2016). Consequently, IMOs derived from coconut waste can enhance the nutritional quality of coconut pulp as a feed ingredient. This study aimed to evaluate the nutritional quality of coconut pulp using IMOs.

Material and Methods

The initial stage of this research involved producing IMOs from coconut water and coconut pulp at a ratio of 1 liter of coconut water to 1 kg of coconut meal, which was fermented anaerobically for 10 days. The resulting IMOs were analyzed for color, aroma, pH, total bacteria, cellulase, and mannanase activities. Subsequently, IMOs were applied to ferment coconut pulp. The study employed a completely randomized design with four treatments and four replications. The treatments consisted of four fermentation durations for coconut meal: 0, 7, 14, and 21 days. The fermentation of coconut pulp utilized indigenous microorganisms (IMOs)

at a dosage of 4%. The parameters observed in fermented coconut pulp are cellulase activity, mannanase activity, and pH. The proximate analysis was conducted to determine crude fiber, crude protein, ether extract, and Nitrogen-Free Extract (NFE) (AOAC, 2005).

The color and aroma of IMOs were observed by visual [Mullagulova et al., 2021]. Total bacteria were determined using the Total Plate Count (TPC) method. Samples were serially diluted and inoculated onto Plate Count Agar (PCA) using the pour plate technique. The plates were incubated at 37°C for 48 hours, and the resulting colonies were counted and expressed as CFU/mL (Li et al., 2020). Lactic acid bacteria were also determined using the Total Plate Count (TPC) method. Samples were serially diluted and inoculated onto de Man, Rogosa, and Sharpe (MRS) agar using the pour plate technique. The plates were incubated anaerobically at 37°C for 48 hours. The resulting colonies were counted and expressed as CFU/mL (Harnentis et al., 2022).

Cellulase Activity

Cellulase enzyme activity was determined using the Nelson method as described by Triani et al. (2024). One milliliter of crude enzyme was mixed with 1 mL of extract (0.5% carboxymethyl cellulose [CMC] dissolved in 10 mL of buffer) and incubated at 40°C for 30 minutes in a water bath shaker. After incubation, 1 mL of Nelson AB solution was added to the mixture, which was then heated in boiling water fofor 20 minutes. After the mixture cooled, 1 mL of phosphomolybdate solution and 7 mL of distilled water were added.er were added. The absorbance was measured using a spectrophotometer at a wavelength of 575 nm, and enzyme activity was calculated based on a standard curve.

Mannanase Activity

Mannanase enzyme activity was measured based on the amount of mannose released using a modification of the Nelson method (1944). One milliliter of substrate (0.5 g of substrate dissolved in 100 mL of 50 mM phosphate buffer at pH 7) was mixed with 1 mL of mannanase enzyme solution and incubated at 50°C for 30 minutes. The reaction was stopped by heating the mixture in boiling water at 100°C for 5 minutes, followed by centrifugation for 5 minutes. One milliliter of the supernatant was added to 1 mL of Nelson reagent (a combination of 25 mL of Nelson A and 1 mL of Nelson B), heated at 100°C for 20 minutes, and cooled to room temperature. Subsequently, 1 mL of phosphomolybdate solution and 7 mL of distilled water were added. The red-brown color intensity was measured using a spectrophotometer at a wavelength of 595 nm.

Proximate Analysis

The crude fiber, crude protein, ether extract and NFE content in fermented coconut pulp were analyzed using the AOAC (2005) methods.

Data Analysis

The characteristics of IMOs were analyzed descriptively and qualitatively, while enzyme activity and the nutritional composition of coconut pulp were analyzed using analysis of variance (ANOVA) followed by Duncan's Multiple Range Test (DMRT), as described by Steel and Torrie (1997).

Results and Discussion

IMOs Characteristics

The characteristics of IMOs derived from the research, including aroma, color, odor, pH, cellulase activity, mannanase activity, and the total bacterial colony, are presented in Table 1. The aroma of the IMOs produced from coconut water and coconut meal has a sour, tapai-like smell, indicating that the MOL has a complete fermentation process

Table 1. Characteristics of IMOs

Characteristics of IMOs	Observation

Aroma	Sour
Colour	Light brown
pH	3.3
Total Bacteria	7.2×10^{12} cfu/ml
Total Lactic Acid Bacteria	5.4×10^{12} cfu/ml
Cellulase Activity	2.1 U/ml
Manannase Activity	7.73 U/ml

The sour odor with a tapai-like aroma in the IMOs suggests that it is mature and ready for application (Yuliana, 2021). The tapai-like sour smell in the IMOs is caused by the hydrolysis of carbohydrates by microorganisms, which produce organic acids such as lactic acid and alcohol (Yunilas et al., 2022). The pH value is a chemical property of the IMOs, and the resulting IMOs have a very low pH of 3.2. The activity of microorganisms, which release H+ ions to form organic acids, also contributes to this pH decrease. The pH of these IMOs is lower than the standard pH range for IMOs produced from fruits and vegetables, which ranges from 3.87 to 4.13 (Swandi et al., 2023).

Total bacterial count in the IMOs produced from coconut water and meal is relatively high, measuring 7.2 × 10^12 cfu/ml, with lactic acid bacteria (LAB) making up a significant portion at 5.4 × 10^12 cfu/ml. This dominance of LAB causes a more acidic MOL with a lower pH. The LAB are group of gram-positive bacteria capable of hydrolyzing carbohydrate substrates into organic acids while producing another metabolite. Organic acids, including propionic, acetic, and lactic acid, create unfavorable environment for spoilage microorganism growth (Bangar et al., 2022)

Cellulase activity is indicated by the activity of cellulolytic bacteria capable of degrading crude fiber. The cellulase activity in the IMOs derived from coconut water and coconut meal is 2.1 U/ml. This amount is considered low if compared to the cellulase activity of cellulolytic bacteria from cassava waste, which reaches 7.58 U/ml (Triani et al., 2024). This variation is due to the high mannose fiber content in coconut pulp, limiting the growth of cellulolytic bacteria in the IMOs. Bacterial growth tends to align with the available substrate (Triani et al., 2024). Although the cellulase enzyme activity in the produced IMOs is relatively low, the mannanase activity is significantly higher at 7.73 U/ml. The mannanase activity observed in this study is higher than that of Aspergillus niger, which has 0.33 U/ml cellulase activity (Bahri et al., 2022) and is considered a known mannanase producer (Dawood et al., 2020).

Cellulase and Mannanase Enzyme Activity, and pH Values in Fermented Coconut Pulp

The effectiveness of IMOS as a bioactivator or for coconut pulp fermentation can be evaluated based on fermentation process and nutritional quality of the fermented coconut pulp. Cellulase and mannanase enzyme activities, along with pH values of fermented coconut pulp, are presented in Table 2, while nutritional quality, including crude fiber, crude protein, and crude fat, is shown in Table 3.

Table 2. Cellulase and mannanase activity and pH value in fermented coconut meal

Treatments	Selulase Activity	Manannase Activity	pH
T0	1.28^{a}	4.62^{a}	4.40^{a}
T1	1.57^{b}	5.19^{a}	3.95^{b}
T2	1.77^{b}	5.09^{a}	3.64^{c}
T3	1.68^{b}	6.99^{b}	3.69^{c}

Note : Description: Different superscripts in the same column indicate significant differences (P<0.05) and very significant differences (P<0.01). T0=control, T1=7 days fermentation, T2=14 days fermentation, T3=21 days fermentation

The results of the study indicate that the fermentation duration of coconut pulp significantly (P<0.01) affects cellulase activity, mannanase activity, and pH values. Cellulase activity on days 7, 14, and 21 was significantly higher (P<0.01) than on day 0. Mannanase activity was highest (P<0.01) at 21 days of fermentation, while the lowest pH values (P<0.01) were observed on days 14 and 21 of fermentation.

The increased cellulase activity on days 7, 14, and 21 resulted from the growth of cellulolytic microbes, which also enhanced the activity of microbes that degrade fibers in coconut pulp. The synthesis and level of activity of cellulolytic enzymes depend on the presence of inhibitors and inducers as well as other physical and chemical parameters. Cellulose with a suitable structure, such as bran and pulp, is an excellent biological inducer for microbiological cellulase (Grata, 2020). The results of the study in Table 2 also show that the optimal fermentation time in coconut pulp using MOL is 14 days. When the fermentation duration is increased to 21 days, cellulase activity tends to decrease; however, this change is not statistically significant. This decrease is attributed to an increase in inhibitors that reduce cellulase activity, which occurs alongside the rise in cellulose hydrolysis products. Cellulose hydrolysis products are most often cellulase inhibitors; cellobiose inhibits endoglucanase and exoglucanase activity, while the presence of glucose can suppress β-glucosidase activity. Other compounds, including solvents, phenolic compounds, and surfactants, can also be inactivators (Grata, 2020). The value of cellulase enzyme activity in this study was relatively low; the decrease was due to the high mannan content in coconut pulp being an inhibitor of cellulase activity (Kumar et al., 2014).

Mannanase activity in coconut pulp increased significantly after 21 days of fermentation, whereas the activity observed at 7 and 14 days of fermentation did not differ significantly from the control, suggesting optimal mannanolytic microbial activity at 14 days of fermentation. Enzyme activity, including mannanase, depends on the growth and development of microorganisms, each of which has a specific growth period (Triani et al., 2024). Microorganisms are an important source of mannanase, an enzyme that exhibits catalytic activity. Mannanase activity will degrade feed ingredients rich in mannan. β-Mannanase, the primary mannan-degrading enzyme, hydrolyzes β-1,4-mannoside bonds in mannan (Wang et al., 2024).

The acidity of the fermented product serves as a gauge for the effectiveness of the fermentation process. A low pH, classified as acidic, will prevent the growth of pathogenic and spoilage microbes, resulting in a sour aroma, which is characteristic of fermented products. The study findings showed that the pH value of coconut pulp fermentation after 14 and 21 days was lower compared to other treatments. This decrease is attributed to the continued microbial growth and increased activity until day 14, which led to higher production of organic acids as secondary metabolites, thereby lowering the pH value. The duration of fermentation significantly influences the pH value, as it fosters the growth of microbes that generate organic acids (Sandi et al., 2023). The pH value of fermented coconut pulp was lower than that of cassava waste fermentation, which had a pH range of 4.28–4.25 (Triani et al., 2024).

Nutritional Quality of Fermented Coconut Pulp

The nutritional quality of fermented coconut pulp also reflects the effectiveness of IMOs as a bioactivator. Table 3 presents the nutritional composition, which includes crude fiber, crude protein, and crude fat. The results of this study show that fermentation duration significantly affects (P<0.01) crude fiber, crude protein, and crude fat in coconut pulp.

Table 3. Crude Fiber, Crude Protein, Ether Extract and NFE of Fermented Coconut Meal

Treatments	Crude Fiber (%)	Crude Protein (%)	Ether extract(%)	NFE (%)
T0	28.03^{a}	4.84^{a}	35.04^{a}	28.97^{a}
T1	22.01^{b}	5.37^{b}	26.64^{b}	36.76^{b}
T2	18.47^{c}	7.01^{c}	24.49^{c}	37.65^{b}
T3 (21 Days)	20.92^{b}	4.90^{a}	25.23^{c}	33.12^{c}
P-value	$2.72x10^{-8}$	$1.49x10^{-8}$	$2.84x10^{-7}$	$1.05x10^{-8}$

Note : Description: Different superscripts in the same column indicate significant differences (P<0.05) and very significant differences (P<0.01). T0=control (0 days fermentation), T1=7 days fermentation, T2=14 days fermentation, T3=21 days fermentation.

Coconut pulp fermented for 14 days exhibited significantly lower (P<0.01) crude fiber and ether extract [R1] content, alongside the highest crude protein (P<0.01). The crude fiber content of coconut pulp was lower after 14 days of fermentation, measuring 18.47%. This fermentation treatment reduced crude fiber by 34.10% compared to the control (unfermented coconut pulp), which had a crude fiber content of 28.03%. The reduction in crude fiber content in fermented coconut pulp is due to the increased activity of cellulase enzymes, which degrade fiber components such as cellulose into glucose. Enzyme activity is influenced by fermentation duration and microbial growth, highlighting the significant role of time in the microbial degradation of cellulose (Yin et al., 2024).

The crude protein content in coconut pulp. The crude protein content increased significantly from 4.84% before fermentation (control) to 7.01% after 14 days of fermentation, representing a 44.63% increase, which is attributed to the rising microbial population.g microbial population, as indicated by the enhanced enzymatic activity. A high microbial population during fermentation contributes to increased protein content through protein synthesis within the microbial cells, particularly bacteria. Protein synthesis is the process of forming polypeptide compounds within cells, which supports bacterial growth and activity, thereby enhancing protein content (Renaldi et al., 2023).

Extending the fermentation period to 21 days resulted in a decrease in crude protein content, returning to 4.90%, which is comparable to the control treatment. This decline is attributed to a reduction in microbial population during fermentation due to the depletion of available nutrients. Microbial growth is influenced by the availability of nutrients (Kumakura et al., 2023). Microorganisms, as unicellular entities, contribute to the crude protein content of the feed; therefore, a decrease in bacterial population directly leads to a reduction in the protein content of fermented coconut pulp.

Coconut pulp is an agricultural product with a high-fat content, and fermentation is expected to reduce its fat levels. The ether extract content of coconut pulp fermented with IMOs decreased significantly as the fermentation duration increased. The lowest crude fat content was observed in coconut pulp fermented for 14 and 21 days, showing a reduction of 30.11%. The decrease in ether extract during fermentation is attributed to the hydrolysis of fats by microorganisms, breaking them down into fatty acids and glycerol. This finding is consistent with the findings of Hayati (2018), who reported that microorganisms in IMOs produce lipase, which hydrolyzes fat into fatty acids and glycerol. Microorganisms subsequently utilize the hydrolysis products for their growth.

Fermentation duration significantly affected (P<0.01) the nitrogen-free extract (NFE) content of coconut pulp. Fermentation for 7 and 14 days resulted in the highest NFE content (P<0.01) at 36.76% and 37.65%, respectively. The elevated NFE content was attributed to the activity of cellulase and mannanase enzymes in degrading fiber components into glucose and mannose.

The cellulase enzymes present in IMOs degrade crude fiber in coconut pulp by hydrolyzing and breaking the glycosidic bonds in cellulose, resulting in cellobiose and glucose (Machiel et al., 2023). Meanwhile, mannanase enzymes hydrolyze the glycosidic bonds in mannan chains, breaking down polysaccharides to mannose products. Furthermore, Mannan can be sustainably and efficiently degraded into mannooligosaccharides (MOS), which are important as prebiotics (Capeti et al., 2023).

This was evidenced by the increased cellulase activity alongside a decrease in crude fiber content and the upward trend in mannanase activity, albeit not statistically significant. However, extending the fermentation period to 21 days led to a significant decline in NFE content despite the increase in mannanase activity. This decline is likely due to the utilization of mannose, a hydrolysis product, by microbes as a nutrient source, as the nutrient availability in the fermentation medium diminishes over time. Microbes consume a portion of the fiber degradation products for their growth. Thus, with prolonged fermentation, glucose and mannose are increasingly utilized by microbes as the nutrients in the medium become progressively diminished (Triani et al., 2024). This study is in line with Hindratiningrum (2023), who stated that the fermentation period of cassava leaves using EM4 for 14 days resulted in an NFE of 37.93%.

Conclusions

The IMOs derived from coconut water and coconut meal have the potential to be utilized as a bioactivator for fermentation to enhance the nutritional quality of coconut pulp. Fermenting coconut meal for 14 days yielded optimal nutritional quality with lower crude fiber and ether extract contents, as well as increased crude protein and nitrogen-free extract (NFE) levels.

References

AOAC. 2005. Official Methods of Analysis of the Association of Analytical Chemist. In Virginia USA: Association of Official Analytical Chemist, Inc.

Bahri S, B Sundu and MR Aprianto. 2019. Mannanase activity produced through fermentation of coconut flour at various pH by *Aspergilus niger*. International Seminar on Science and Technology 2018 IOP Conf. Series: Journal of Physics: Conf. Series 1242 (2019) 012009 IOP Publishing doi:10.1088/1742-6596/1242/1/012009.

Bangar SP, S Suri, M Trif and M Trif. 2022. Organic acids production from lactic acid bacteria. Food Bioscience. 46: 101615. https://doi.org/10.1016/j.fbio.2022.101615.

BPS. 2024. Food Crop Statistics. Badan Pusat Statistik, Jakarta.

Capetti CCM, ANG Dabul, VOA Pellegrini, I Polikarpov. 2023. Manannases and othen mannan-degrading enzymes. ScienceDirect. 12:279-293. https://doi.org/10.1016/B978-0-323-91805-3.00013-7.

Dawood A and K Ma. 2020. Applications of Microbial β-Mannanases. Bioeng Biotechnol. 15(8) : 598630. http:// doi: 10.3389/fbioe.2020.598630.

Grata K. 2020. Determining cellulolytic activity of microorganisme. Chem Didact Ecol Metrol. 25(1) :133-143. http://dx.doi.org/10.2478/cdem-2020-0010.

Hasanah A, S Rahayu, B Hartoyo, FM Suhartati, Munasik and N Hinratiningrum. 2024. Optimization of Hydrolytic Enzyme Production from Tempeh Starter to Increase In Vitro Pepsin Digestibility of Black Soldier Fly Maggot. Animal Production. 26 (2):112-124. https://doi.org/10.20884/1.jap.2024.26.2.278.

Harnentis, Y Marlida, YS Nur, Wizna, MA Santi, N Septiani, F Aldzetlty and N Huda. 2020. Novel probiotic lactic acid bacteria isolted from indegenous fermented food from West Sumatera Indonesia. Vetrinary Word. 13(9):1922-1927. http://dx.doi.org/10.14202/vetworld.2020.1922-1927. Hidayati SG. 2017. Pemanfaatan Mikroorganisme Lokal Sebagai Inokulum untuk Meningkatkan Kualitas Ampas Kelapa Sebagai Bahan Pakan Alternatif Ayam Buras Periode Pertumbuhan. Disertasi. Pascasarjana, Universitas Andalas. Padang.

Hindratiningrum N, R Fitria and SA Santosa. 2023. The Nutrition Quality of Cassava Leaf Silage with Different Fermentation Lengths. Animal Production. 25 (3): 179-185. https://doi.org/10.20884/1.jap.2023.25.3.222.

Khasanah H, L Purnamasari and DE Kusbianto. 2020. Pemanfaatan MOL (Mikroorganisme Lokal) sebagai substitusi biostarter EM4 untuk meningkatkan mualitas nutrisi pakan fermentasi

berbasis tongkol dan tumpi jagung. Prosiding Seminar Nasional Teknologi Peternakan dan Veteriner.Pp 345-352. DOI: http://dx.doi.org/10.14334/Pros.Semnas.TPV-2019-p.345-352. Kumar R and CE Wyman. 2014. Strong cellulase inhibition by mannan polysaccharides in cellulose Conversion to Sugars. Biotechnology and Bioengineering. 111(7): 1341 - 1353.

Laksono J, T Karyono and H Haniati. 2023. Nilai nutrisi ampas kelapa (*Cocos nucifera L.)* yang difermentasi menggunakan *Aspergillus niger* dengan waktu berbeda sebagai ransum ternak unggas. Jurnal Ilmu dan Teknologi Peternakan Indonesia. 9(1): 42-48.

Li S, S Tang, Q He, J Gong and J Hu. 2020. Physicochemical, textural and volatile characteristics of fermented milk co-cultured with Streptococcus thermophilus, Bifidobacterium animalis or Lactobacillus plantarum. International Journal of Food Science & Technology. 55(2): 461–474. https://doi.org/10.1111/ijfs.14279.

Maciel C, DSP Rosinelson, DDN Lidiane, ADO Taciane, CAC Gilson and SL Alesandra. 2023. Health Exposure Risks and Bioremediation of Cyanide in Cassava Processing Effluents: An Overview. Journal of Water Process Engineering 55 : 104079.

Mullagulova GM, OV Avtuyhova, YM Rebezov, DI Vorobyev, and AV Goncharov. 2021. The results of organoleptic assessment of a fermented milk product for functional nutrition. IOP Conference Series: Earth and Environmental Science. 677(3): 032041. https://doi.org/10.1088/1755-1315/677/3/032041.

Patel N and S Gautam. 2024. Microbial β-mananase enzyme: a depth review based on properties and applications. IJRTI. 9(4): 52-58.

Steel, R. G. D., and J. H. Torrie. 1997. *Principles and Procedures of Statistics: A Biometrical Approach*. 3rd ed. New York: McGraw-Hill.

Sundu B, U Hatta, S Mozin, N Toana, Hafsah, Marhaeni and S Sarjuni. Coconut meal as a feed ingredient and source of prebiotic for poultry. 2020. The 2nd International Conference of Animal Science and Technology IOP Conf. Series: Earth and Environmental Science 492: 012126. doi:10.1088/1755-1315/492/1/012126.

Triani HD. 2024. Optimalisasi Limbah Ubi Kayu Melalui Bioteknologi Sebagai Bahan Pakan Lokal Untuk Peningkatan Produktivitas Itik Kamang Pedaging. Disertasi. Pascasarjana, Universitas Andalas. Padang.

Triani HD, Y Marlida, A. Yuniza, Husmaini and WD Astuti. 2024. Isolation and characterization of cellulose and cyanide degrading bacteria from cassava waste as inoculants in feed fermentation. International Journal of Veterinary Science 13(3): 384-390; https://doi.org/10.47278/journal.ijvs/2023.110.

Wang P, X pei, W Zhou, Y Zhao, P Gu, Y Li and J Gao. 2024. Research and aplication preogress of microbial β-manannase. World journal og microbiology and biotechnology. 40 : 169. https://doi.org/10.1007/s11274-024-03985-1.

Yin X, Y Bi, J Niu, W Shao and G Liang. 2024. Metabolic pathway analysis of the biodegradation of cellulose by Bacillus velezensis. Heliyon. 10(19) : e38375. https://doi.org/10.1016/j.heliyon.2024.e38375 .

Yuliana M. 2021. The effect of local microorganism (MOL) as liquid organic fertilizer to the growth of Ipomea reptans Poir. J. Biota. 7(1): 51-56.

Yulianto R, G Meriana and L Nashrullah. 20200. Degradation fermented feed animal based local mikroorganism to increased nutrient quality of the waste gambir fermented. Prevenire. 1(3):153-160.

Yunilas, AZ Siregar, E Mirwhandhono, A Purba N Fati and T Malvin. 2022. Potensi dan karakteristik larutan mikroorganisme lokal (MOL) berbasis limbah sayur sebagai bioaktivator dalam fermentasi. J. of Livestock and Animal Health. 5(2): 53-59.

Yunilas, N Ginting, TH Wahyuni, M Zahoor, N Fati and A Wahyuni. 2021. Effect of Various Doses of Local Microorganism Additives on Silage Physical Quality of Corn (*Zea mays L.)* Waste. Sarhad Journal of Agriculture. 37(1):197-206. . https://dx.doi.org/10.17582/journal.sja/2022.37.s1.197.206.