



Study of Noni Fruit Extract Microcapsule on Enzymes Activity, Digestibility and Hematologic of Sentul Chicken

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Abstract | The active compounds in noni fruit can increase the efficiency of digestion and the utilization of nutrients in tissue cells. This present work aims to determine the optimum level of microcapsules of noni fruit extract (MFNE) in the diet to improve digestibility, digestive enzyme activity, and hematological indicators in Sentul chickens (SC). This study involved a 14-week-old female chicken that was maintained in cages until it reached 24 weeks of age. We employed a Completely Randomized Design (CRD) consisting of five different treatments and four replications, with each replication including two chickens. The addition of MFNE products at a concentration of 225 mg/kg to the feed positively influenced both dry matter digestibility and crude protein digestibility. Furthermore, it significantly affected the levels of amylase and protease activity; however, there was no significant effect on lipase activity. Treatment with 150 to 225 mg/kg significantly improved hematological indicators. It was concluded that 150 mg of the MFNE product could be used as a feed additive in poultry to replace Antibiotic Growth Promoters (AGPs).

Keywords | Noni fruit, Microcapsule, Digestive, Hematology, Chicken, Metabolism

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INTRODUCTION

SC is a dual-purpose local chicken known for producing both meat and eggs. It has the potential to be developed as a reliable source of animal protein for the community. The advantages of SC compared to other native chickens include higher productivity in both meat and eggs, faster weight gain, and improved disease resistance. One effort to improve production performance is the provision of feed additives. Feed additives in the ration component are essential in optimizing animal productivity.

Antibiotic growth promoters (AGPs), such as certain additives used in poultry feed, have been restricted due to their potential to create harmful contaminants in animal products, posing risks to consumer health. One possible substitute for AGPs is noni fruit (*Morinda citrifolia*). This fruit, derived from a tropical plant, may effectively replace AGPs in animal diets because it is rich in antioxidants, phenolic compounds, and flavonoids. Krishnaiah *et al.* (2015); Singh *et al.* (2006) showed that noni fruit contains several antibacterial compounds, including saponins, alizarin, anthraquinones, aucubin, and scopoletin (Mushawwir *et al.*, 2024).

Some antioxidant compounds found in noni fruit include pyridine-3-carboxamide, oxime, n-(2-trifluoromethyl phenyl), beta-sitosterol, n-hexadecanoic acid, and squalene (Sogandi and Nilasari, 2019). Other active components are flavonoids, anthraquinones, tannins, acubin, alkaloids, alizarin, and triterpenes (Purwanti *et al.*, 2024). Nugraheni *et al.* (2017) and Diarra *et al.* (2019) stated that anthraquinone compounds can inhibit the growth of several bacteria, including *Staphylococcus aureus*, *Bacillus subtilis*, *E. coli*, *Salmonella*, and *Shigella dysenteriae* and can increase digestibility, lower cholesterol, modulate the immune system, and reduce heat stress in poultry. Furthermore, anthraquinones can influence the pH of the digestive tract, making it more acidic. This acidity allows protein-degrading enzymes to function optimally, enhancing protein absorption by the body (Widianto *et al.*, 2015).

Noni fruit is rich in crude fiber and contains anti-nutrients, so it must be processed before being used as a feed additive. One effective processing technique is extraction, specifically the maceration method. Additionally, encapsulation methods should be employed to protect the active compounds from unfavorable conditions (Hasrini *et al.*, 2017).

Encapsulation technology can enhance the utilization of anthraquinone compounds derived from noni. The addition of MFNE (Morbidity-Free Noni Extract) containing anthraquinones ensures that these compounds do not undergo structural damage in the stomach, allowing for optimal absorption in the small intestine. The small intestine is crucial for the absorption of feed nutrients, enabling anthraquinone compounds to be digested and absorbed more effectively. Furthermore, the anthraquinone compounds in MFNE can help prevent and improve the villous structure of the small intestine, thereby increasing the rate of nutrient absorption. Additionally, these compounds suppress the growth of pathogenic microbes in the intestine (Velmurugan and Citarasu, 2010).

Biochemically, these conditions promote the growth of absorptive cells in the digestive tract. This enhancement, in turn, leads to better biochemical conditions, supported by degrading enzymes such as proteases, lipases, and amylases. As a result, digestion and absorption of carbohydrates, proteins, and lipids are significantly improved (Nguru *et al.*, 2022). The alkaloid compounds xeronine and proxeronine in MFNE stimulate nutrient metabolism, activate inactive proteins, and repair the structure and function of cellular proteins (Kharazi *et al.*, 2022; Mushawwir *et al.*, 2024; 2021a, b). The proxeronine enzyme in the small intestine will convert proxeronine into xeronine, then xeronine will modify the molecular structure of proteins so that the absorption process of food substances becomes better and faster (Djauhariya *et al.*, 2016; Mushawwir *et al.*, 2023).

The mechanism by which flavonoid compounds function as antimicrobial agents involves damaging the bacterial cell wall, specifically the peptidoglycan layer. This damage occurs through diffusion, leading to lysis and ultimately causing bacterial cell death. Flavonoids act as antibacterials by inhibiting cellular functions, as they form complex compounds with extracellular proteins that harm bacterial cells. This process results in the release of intracellular compounds from the bacteria (Firmansyah *et al.*, 2024). Controlling the bacterial population in the digestive system through flavonoid compounds can enhance chicken performance. Additionally, antioxidant compounds present in noni fruit extracts can convert free radicals into more stable forms, preventing chain reactions that result from free radical damage. This protective effect can positively influence the growth rate of chickens (Zabolil *et al.*, 2013; Mushawwir *et al.*, 2022a).

The essential oil compounds in noni fruit can also improve the work of poultry digestive organs by stimulating the gallbladder wall to secrete bile and stimulate the release of pancreatic sap containing amylase, lipase and protease enzymes that are useful for improving the digestion of feed ingredients such as carbohydrates, fats and proteins. The alkaloid content of proxeronin in noni fruit is quite large and will be converted into xeronin with the help of the enzyme proxeroninase. In addition, xeronine leads to the dilation of the small intestine; it also improves the function of the thyroid gland and thymus, which are essential for immunity and resistance to external infections, activates enzymes, and regulates the function of proteins in cells (Rahmania *et al.*, 2022; Mushawwir *et al.*, 2022b; Manin *et al.*, 2024; Setiawan *et al.*, 2024).

Research reported by Adriani *et al.* (2017) showed that the use of noni fruit flour added to the ratio of layer phase quail with a concentration of 0.75% was able to maintain the number of erythrocytes, hemoglobin levels, and hematocrit values of quail. Several other studies have shown that the provision of noni can reduce lipid profiles in quails; according to the results of research by Rahmania *et al.* (2022), the provision of 1.2% noni extract supplemented with 60 mg Cu and 360 mg Zn can reduce blood cholesterol to 111.83 mg/dL. Adriani *et al.* (2018) stated that giving 0.3% noni plus 4% palm sugar can lower cholesterol to 91 ± 2.83 mg/dL, triglycerides to 12 ± 2.83 mg/dL, and low-density lipoprotein (LDL) to 11.6 ± 0.85 mg/dL, and giving 0.3% noni plus 3% palm sugar can reduce high-density lipoprotein (HDL) to 86.5 ± 14.85 mg/dL.

In previous studies, Rindiany *et al.* (2022) demonstrated that administering microcapsules of noni fruit extract (MFNE) at a dose of 125 mg/kg can reduce triglyceride levels to 49.27 mg/dL. Additionally, a dose of 250 mg/kg of MFNE resulted in the lowest cholesterol levels recorded

at 114.84 mg/dL. Numerous studies have highlighted the benefits of noni fruit in preventing various diseases and its positive effects on human metabolism as well as in multiple animal species. However, there is a lack of research specifically focused on the impact of noni fruit on the enzyme activity of native chickens, particularly in relation to reducing the need for antibiotics. Given this context, utilizing MFNE products as feed additives could be a viable solution for enhancing the immune system and productivity of native chickens while also improving the quality of their meat and eggs.

MATERIALS AND METHODS

Forty female chickens of the SC breed, aged 14 weeks, were raised until they reached 24 weeks. They had an average initial weight of 785.6 grams, with a coefficient of variation of 14.6%. The experimental chickens were assigned to five treatments, each repeated four times, with two chickens per replication. A total of 40 cages were utilized for the study, with each cage measuring 40 cm in length, 35 cm in width, and 30 cm in height. The diet consisted of a mixture of concentrate, corn, bran, and MFNE. Details regarding the nutrient and metabolic energy content, as well as the ration formulation, can be found in [Tables 1 and 2](#).

Table 1: Composition of basal rations (BR).

Feed Ingredients	Quantity (%)
Rice Bran	25.00
Yellow Corn	50.00
Laying Chicken Concentrate	25.00
Total	100

Table 2: Nutrient content and metabolic energy of BR.

Nutrient content	Total
Metabolic energy (kcal/kg)	2777
Crude protein (%)	15.80
Crude fat (%)	5.70
Crude fiber (%)	6.25
Calcium (%)	3.10
Phosphor (%)	0.21
Lysine (%)	0.86
Methionine (%)	0.42

The encapsulation process was modified as follows: the thick noni fruit extract was diluted with distilled water in a 1:1 ratio. Similarly, the maltodextrin coating was diluted with distilled water at the same 1:1 ratio. After dilution, the extract was combined with the maltodextrin coating in a ratio of 70% extract to 30% maltodextrin until the mixture was uniform. The resulting mixture was then dried using the spray drying method, with an inlet temperature

of 125°C and an outlet temperature of 60°C ([Mishra et al., 2014](#)). The composition of the experimental diet and the addition of MFNE and Zinc Bacitracin, in each treatment, is shown in [Table 3](#).

Table 3: Composition of feed ingredients in treatment rations.

Nutrien	Treatments				
	P0	P1	P2	P3	P4
Basal Ration (BR) (%)	100	100	100	100	100
MFNE Products (mg/kg)	0	0	75	150	225
Zinc Bacitracin (mg/kg)	0	50	0	0	0

DIGESTIVES ENZYME ANALYSIS

At 20 weeks of age, intestinal samples were obtained to determine digestive enzyme activity. This examination was conducted at the Faculty of Animal Husbandry test laboratory at Padjadjaran University.

HEMATOLOGIC AND PLASMA CHEMISTRY ANALYSIS

Hematologic and blood chemistry levels, including plasma glucose and cholesterol, were analyzed by collecting blood samples from a pelvic vein at the end of the study using an EDTA project. All analytical procedures were conducted according to the instructions provided with the Biolabo kit (UK).

NUTRIENT DIGESTIBILITY ANALYSIS

The determination of nutrient digestibility was conducted using the following method: Sampling took place in the twenty-fourth week. The sample chickens were fed and had access to drinking water ad libitum. They were fed for 12 hours prior to slaughter. After this period, the chickens were cut open at the breast, and the intestines were removed for fecal sample collection. The fecal samples were dried and analyzed to determine their lignin content, crude protein, dry matter, and organic matter.

STATISTICAL ANALYSIS

Statistical analyses were performed using SPSS software (IBM SPSS version 21). One-way analysis of variance (ANOVA) was applied to determine the effect of treatment. The differences were considered significant at the 0.05 level. In addition, Duncan's test compared different treatments in different ways.

RESULTS AND DISCUSSION

THE ACTIVITY OF DIGESTIVE ENZYMES

The impact of adding MFNE on the activity of digestive enzymes is shown in [Table 4](#). [Table 4](#) demonstrates that the activities of the enzymes protease, amylase, and lipase varied across different treatments. The chicken groups re-

ceiving the P1 and P2 treatments, which involved the addition of MFNE at doses of 75 to 150 mg/kg, displayed higher protease and amylase enzyme activities compared to those treated with zinc bacitracin. Protease enzymes are essential for protein synthesis; they catalyze proteins into amino acids, which are then absorbed into the bloodstream and transported to body cells for the formation of growth tissues. Higher protease enzyme activity correlates with an increased ability of chickens to utilize protein, positively impacting muscle tissue growth.

Table 4: Observations on enzyme levels in the intestine.

Treatments	Protease (U/MI)	Amylase (U/MI)	Lipase (U/MI)
P0	11.97	23.13	3.17
P1	7.08	20.81	2.50
P2	11.27	30.23	3.33
P3	12.20	32.09	3.50
P4	9.28	30.64	4.08

Essential oil compounds found in noni fruit can enhance the function of poultry digestive organs by stimulating the gallbladder wall to secrete bile. This process facilitates the release of pancreatic secretions containing enzymes such as amylase, lipase, and protease, which are beneficial for the digestion of feed ingredients like carbohydrates, fats, and proteins.

The results of the current study indicate that the levels of lipase enzyme were higher in treatments that included MFNE compared to those with Zinc Bacitracin. The addition of noni fruit extract to the diet can enhance bile secretion and improve the functioning of digestive organs, particularly the small intestine. Increased bile secretion promotes lipid absorption and enhances the pancreas's metabolic performance (Mushawwir *et al.*, 2024). Noni fruit extract contains active compounds, specifically flavonoids and alkaloids (Kharazi *et al.*, 2022). Flavonoids have the ability to inhibit lipase enzyme activity, which can lead to an increased excretion of fat through feces.

BLOOD HEMATOLOGY AND BLOOD BIOCHEMISTRY

Table 5 shows the average hematologic level and blood biochemistry values of SC fed the antibiotics zinc bacitracin and MFNE.

The average number of erythrocytes in SC in the P0, P1, P2, and P3 treatments is still in the normal range. This is based on the statement of Iriyanti and Soedirman (2015), which states that the average number of erythrocytes in SC in normal conditions is $2.0 - 3.0 \times 10^6/\text{mm}^3$. The results of Duncan's multiple range test in Table 5 showed that the number of erythrocytes in the P0 treatment was significantly lower than P1, P2, P3, and P4, while P1, P2, and

P3 were not significantly different. Adding MFNE up to 225 mg/kg in the diet can maintain the erythrocyte count of SC in the normal range. The results of this study indicate that flavonoid activity contained in MFNE can affect blood synthesis. Flavonoids can improve the performance of blood-producing organs so that blood production increases (Wahjuningrum *et al.*, 2008). Flavonoids in the blood stimulate the kidneys to secrete glycoprotein hormones that stimulate increased erythrocyte cell formation.

Table 5: Average hematology and biochemistry values of SC blood.

Parameters	Treatments				
	P0	P1	P2	P3	P4
Hematologic					
Number of Erythrocytes (million/mm ³)	2.70 ^a	3.52 ^b	3.11 ^b	3.33 ^b	3.01 ^b
Hematocrit Level (%)	26.75 ^a	26.50 ^a	28.00 ^a	24.00 ^a	26.00 ^a
Hemoglobin level (g/dL)	11.99 ^a	13.37 ^a	12.64 ^a	13.21 ^a	13.35 ^a
Blood Biochemistry Status					
Blood cholesterol (mg/dL)	166.33 ^a	166.57 ^a	159.88 ^a	158.21 ^a	148.32 ^a
Blood triglycerides (mg/dL)	103.89 ^a	104.05 ^a	100.07 ^a	87.75 ^b	81.48 ^b
Glucose (mg/dL)	124.94 ^a	126.09 ^a	126.81 ^a	126.33 ^a	125.44 ^a

Different superscript letters in the same column indicate significantly different effects ($P < 0.05$).

Microencapsulation protects bioactive or phenolic compounds such as flavonoids from environmental influences such as temperature, light, and oxygen. The hemoglobin levels obtained are within the normal range of 11.99 - 13.37 (g/dl), and the hematocrit value is still within the normal range of around 22-35%. This is due to the active compound xeronine and other antioxidants in noni fruit extract. Antioxidants prevent changes in the structure and function of cell membranes. Xeronine activates and repairs damaged hemoglobin so that it can work normally.

The results of the analysis of variance indicated that the administration of noni fruit extract microcapsules had a significantly different effect ($P \leq 0.05$) on the blood triglyceride levels of the subjects. However, it did not show a significant impact ($P > 0.05$) on blood cholesterol levels or blood glucose levels. The observed decrease in blood triglyceride levels in chickens treated with microencapsulated noni fruit extract (MFNE) can be attributed to the presence of alkaloid compounds. These alkaloids reduce triglyceride levels by inhibiting the activity of pancreatic lipase enzymes, which in turn increases fat excretion through feces. As a result, fat absorption by the liver is inhibited,

preventing conversion into cholesterol. This inhibition of pancreatic lipase activity leads to fewer triglyceride deposits in the small intestine since the enzyme typically converts triglycerides into two monoglycerides and two free fatty acids for absorption into the bloodstream.

FEED DIGESTIBILITY

Feed digestibility is a key indicator of feed quality. When feed ingredients have higher digestibility, it signifies that the feed is of good quality for animal consumption and supports better animal production. Research data on the effects of microcapsules containing noni fruit extract on dry matter digestibility (DMDi), crude protein digestibility (CPDi), and organic matter digestibility (OMDi) are presented in Table 6.

Table 6: Effect of MFNE on dry matter digestibility, crude protein digestibility, and organic matter digestibility of SC.

Treatments	Parameters		
	DMDi (%)	OMDi (%)	CPDi (%)
P0	66.41 ± 1.48 ^a	70.28 ± 1.27 ^a	65.57 ± 2.71 ^a
P1	72.86 ± 1.30 ^b	76.46 ± 1.24 ^b	67.23 ± 1.43 ^b
P2	73.94 ± 1.08 ^b	77.31 ± 1.11 ^b	68.13 ± 1.48 ^b
P3	78.09 ± 0.88 ^c	79.11 ± 0.82 ^c	74.74 ± 1.05 ^c
P4	78.02 ± 1.04 ^c	78.46 ± 1.05 ^c	76.63 ± 1.46 ^c

Different superscript letters in the same column indicate significantly different effects ($P < 0.05$).

The results of the statistical analysis indicated that the percentage of dry matter digestibility (KCBK) was significant ($p < 0.05$). This suggests that the administration of MFNE has a positive effect on dry matter digestibility (DMDi). The findings of this study demonstrate that noni fruit is a valuable photogenic plant that can be used as a feed additive due to its rich content of essential compounds. Numerous studies have reported that phytogetic feed additives can enhance nutrient digestibility in the digestive tract and increase carcass weight in broiler chickens (Syed *et al.*, 2021; Rahmania *et al.*, 2022; Purwanti *et al.*, 2024; Firmansya *et al.*, 2024). Therefore, incorporating MFNE into the diet leads to improved nutrient absorption in the digestive tract and a higher digestion value. According to Herath *et al.* (2020) and Adriani *et al.* (2024), over 70% of nutrient absorption occurs in the small intestine, which is influenced by the physiological condition of the duodenal mucosa.

The results indicated that MFNE significantly influenced crude protein digestibility ($p < 0.05$). The diet with the most substantial effect on crude protein digestibility was P4, with a digestibility percentage of 79.63%. This was followed by P3 at 73.74%, P1 at 69.23%, P2 at 64.13%, and P0 at 61.57%. According to Duncan's test results, P0 and P2 significantly differed from P1, P3, and P4. Noni fruit contains several key constituents, including alkaloids, tan-

nins, flavonoids, and carbohydrates. The increase in MFNE levels positively affected the crude protein digestibility of the ration, primarily due to the accumulation of the active compound xeronine as more noni fruit extract was added. Heinecki (1985) and Tanuwiria *et al.* (2022) state that the alkaloid identified is proxeronine, a precursor of xeronin (Sanni and Fatoki, 2017; Mushawwir *et al.*, 2022b). Thus, The role of xeronine is essential for optimizing metabolism. Choi *et al.* (2021) noted that xeronine is a significant alkaloid involved in cell regulation. The high level of CPDi in P3 is attributed to the strong association with the OMDi ratio, which is generally better than that of other treatments. Ration protein digestibility is directly proportional to the digestibility of dry matter and organic matter (Rambet *et al.*, 2015; Nurfauziah *et al.*, 2024). This implies that serotonin found in Noni fruit can promote anabolism, specifically muscle protein synthesis.

The addition of MFNE to the ratio of SC, at various levels, increased the OMDi by 150 to 225 mg/kg compared to the control treatments P0 and P1. MFNE is the most effective treatment for improving nutrient absorption, as the anthraquinone compounds in MFNE can lower intestinal pH. This action inhibits the growth of pathogenic bacteria in the digestive tract and promotes an increase in organic acid concentration due to enhanced growth of lactic bacteria. As a result, there is an improvement in the uptake of organic matter, including proteins, carbohydrates, and lipids. Wang *et al.* (2021) stated that anthraquinone compounds increase intestinal acidity. Lower intestinal pH stimulates higher protease activity. This protease catalysis increases amino acid absorption, improving muscle tissue growth.

CONCLUSIONS AND RECOMMENDATIONS

Incorporating MFNE into the diet at 225 mg/kg significantly improved dry matter digestibility and crude protein digestibility. It notably influenced the activity levels of amylase and protease, but had no significant effect on lipase. 150 mg MFNE can be added to feed additives in SC to replace Antibiotic Growth Promoters (AGPs).

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NOVELTY STATEMENT

Antibiotics have been commonly used as growth promoters

in local chickens, raising concerns among consumers about antibiotic residues. However, noni extract as a feed additive represents a significant breakthrough for local chickens. Its effectiveness may surpass that of antibiotics, particularly in stimulating various mechanisms within the tissues of local chickens.

AUTHOR'S CONTRIBUTIONS

All authors contributed equally during this article's research preparation, conduct, and writing.

CONFLICT OF INTEREST

The authors have declared no conflict of interest.

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