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PJBS

ISSN 1028-8880

Pakistan Journal of Biological Sciences

ANSInet
Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

OPEN ACCESS Pakistan Journal of Biological Sciences

ISSN 1028-8880
DOI: 10.3923/pjbs.2025.685.690

Research Article
Effect of Noni Fruit Extract Microcapsules on the Profile and Biochemistry of Sentul Chicken Blood in the Layer Phase

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Abstract

Background and Objective: Noni fruit (*Morinda citrifolia* L.) contains bioactive compounds with the potential to improve lipid metabolism, reduce inflammation and enhance poultry health. However, its application in poultry feed remains underexplored. This study aimed to evaluate the effect of noni fruit extract microcapsules (NFEM) on lipid profile, blood biochemistry and inflammatory markers in Sentul chickens during the laying phase. **Materials and Methods:** A total of 40 hens aged 24 weeks were randomly allocated into five dietary treatments: Control (T1), 50 mg/kg zinc bacitracin (T2), 75 mg/kg NFEM (T3), 150 mg/kg NFEM (T4) and 225 mg/kg NFEM (T5). Blood samples were analyzed for lipid profile, liver function markers and hematological parameters. Data were subjected to Analysis of Variance (ANOVA) and differences among treatments were tested using Duncan's Multiple Range test at a significance level of $p<0.05$. **Results:** Dietary supplementation with NFEM significantly decreased LDL cholesterol levels and increased HDL cholesterol levels ($p<0.05$). A notable reduction in leukocyte and lymphocyte counts indicated lowered inflammation in NFEM-treated groups. Blood biochemical parameters further suggested improved liver function compared with the control. **Conclusion:** Noni fruit extract microcapsules improved lipid metabolism, reduced inflammation and supported liver function in Sentul chickens during the laying phase. These findings highlight the potential of NFEM as a natural feed additive to enhance poultry health and productivity.

Key words: Blood biochemistry, blood profile, feed additive, microcapsules, noni fruit extract, sentul chic

Citation: Adriani, L., T. Widjastuti, A. Mushawwir, R.L. Balia, S. Adil and Mirnawati, 2025. Effect of noni fruit extract microcapsules on the profile and biochemistry of sentul chicken blood in the layer phase. Pak. J. Biol. Sci., 28: 685-690.

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Local chickens cultivated in Indonesia include various types, one of which is Sentul chicken, which comes from Ciamis Regency, West Java. Sentul chicken is included in the dual-purpose category because it has a faster growth rate than other local chickens, making it suitable as a broiler chicken. In addition, with a fairly high level of egg productivity, Sentul Chicken also has the potential to be utilized as laying hens. Sentul chicken production as one of the local poultry in Indonesia, has great potential in supporting food security, especially as a source of animal protein. However, the metabolic efficiency of Sentul chickens in utilizing feed nutrients to support productivity, such as egg production, still requires optimization.

The genetic superiority of Sentul chicken needs to be supported by good maintenance management to achieve maximum production performance. One important aspect in this case is feed management. The addition of certain compounds in feed rations can be a solution to overcome nutritional and metabolic problems in poultry, so that it can improve production performance[1-3]. One alternative to additional feed is by utilizing herbal plants, namely noni fruit. Noni fruit (*Morinda citrifolia* L.) contains various compounds such as alkaloids, flavonoids and antioxidants. These compounds have benefits as antimicrobials and are rich in minerals and vitamins that can improve the immune system, improve appetite and support nutrient absorption in the intestines, thereby increasing ration efficiency[4].

The noni fruit extraction process aims to separate the main compounds from the fruit using the right solvent. Noni fruit extract contains unsaturated organic acid compounds with short chains that are relatively unstable, thus requiring metal catalysts such as copper (Cu) and zinc (Zn) to increase their stability. The mineral copper (Cu) plays a role in the production of red and white blood cells and stimulates the release of iron to form hemoglobin that transports oxygen throughout the body. The presence of copper allows the body to utilize iron, supports nerve function and accelerates bone growth. Zinc (Zn) is a micro mineral that is distributed in animal and plant tissues, plays an important role in metabolism, besides that Zn also helps strengthen the immune system, supports cell growth, increases appetite, hatchability, egg production and supports bone and feather growth[5].

One important aspect of this optimization is the management of blood profiles and biochemistry that play a key role in supporting the physiological functions of chickens, including energy production, cell membrane formation and general health of the birds. Lipoprotein profiles, specifically High-Density Lipoprotein (HDL) and Low-Density Lipoprotein (LDL) levels, are often used as indicators of lipid health in the blood. On the other hand, metabolic stress, inflammation and liver function also contribute to Sentul chicken performance. Systemic inflammation, which can be measured through parameters such as leukocyte, lymphocyte, TNF-α and interleukin (IL) levels, provides an overview of the immune condition of the chicken body[6-8]. This study aims to evaluate the effect of treatments on the concentration of lipid regulators, lipid transport, inflammation and liver health of Sentul chickens in the layer phase. By understanding these relationships, the results of the study are expected to be the basis for designing more efficient feeding strategies, so as to increase the productivity and welfare of local chickens, such as Sentul chickens.

MATERIALS AND METHODS

Study area: This research was conducted at the Poultry House of the Faculty of Animal Husbandry, Padjadjaran University, Sumedang, Indonesia from September to December, 2024.

Animal, housing and location: A total of 40 female Sentul chickens used as research livestock were obtained from the Jatiwangi Poultry Development and Breeding Center (BPPTU) and were intensively raised for 12 weeks at the age of 24 weeks. These Sentul chickens were divided into 5 feed treatment groups with 4 repetitions each, which were randomly placed into 40 cage units. The average initial weight of female Sentul chickens was 1341.1 g with a coefficient of variation of 14%. The treatments given included the level of microencapsulation of mengkudu fruit extract in the ratio, namely:

- T1 = Basal
- T2 = Basal+50 mg/kg zinc bacitracin
- T3 = Basal+75 mg/kg NFEM
- T4 = Basal+150 mg/kg NFEM
- T5 = Basal+225 mg/kg NFEM

Data and blood sample collection: Sampling was carried out at the end of maintenance. Blood samples were taken using a syringe inserted into the wing vein (externa pectoralis vein) as much to 3 mL. Blood samples were put into an EDTA container, then shaken slowly and stored in a cooling box before the blood was analyzed to avoid blood clotting. The blood samples obtained were centrifuged at 3000 rpm for 15 min to obtain blood plasma. Plasma was separated using a micropipette and collected into a 3 mL collection tube. Blood plasma was analyzed using the spectrophotometric technique by mixing reagents and buffer solutions based on the instructions of the Biolabo Kit, France (2015), with a wavelength according to the parameters[9].

Data analysis: The data obtained were analyzed using Analysis of Variance (ANOVA) and treatment differences were evaluated using Duncan's Multiple Range Test (DMRT) at a significance level of p<0.05 with IBM SPSS version 25 software.

RESULTS AND DISCUSSION

Concentration of regulators and lipid transport in Sentul chicken layer phase: The results of this study showed significant changes in the values of the parameters tested, along with the provision of different treatments. The administration of NFEM products had a significant impact on blood cholesterol and triglyceride profiles in Sentul chickens in the layer phase (p<0.05). The study showed that increasing the dose of NFEM can significantly reduce cholesterol levels. The T4 and T5 treatments showed higher HDL (High-Density Lipoprotein) values compared to T1 (control) or T2 (antibiotic control), showing that NFEM can increase HDL up to 25% shown in Table 1.

In the results of the study, PPAR-γ values did not show significant differences (p>0.05) between treatments. The values ranged from 2.03-2.13 µg/dL. This indicates that the treatments given did not significantly affect the expression of PPAR-γ in Sentul chickens in the layer phase. The results showed that the treatment significantly affected the concentration of Apo A1 (p<0.05). The highest value was achieved in treatment T1 (3.97 mg/dL) and the lowest in T5 (2.76 mg/dL). In contrast, Apo A2, which acts as a cofactor in the enzyme lipoprotein lipase (LPL), also showed significant differences (p<0.05). The highest value was achieved in T5 (1.02 mg/dL), while the lowest value was in T2 (0.76 mg/dL). Results showed that Apo B values varied significantly (p<0.05), with the highest value in T1 (0.75 mg/dL) and the lowest in T5 (0.46 mg/dL). The highest value was recorded in T5 (1.13 mg/dL), while the lowest value was in T1 (0.93 mg/dL).

The PPAR-γ is a transcription factor that plays an important role in lipid metabolism and the regulation of insulin resistance. Apolipoprotein A1 (Apo A1) is a major component of HDL that plays a role in cholesterol transportation from tissues to the liver. The decrease in Apo A1 in treatments with certain concentrations indicates that the variation of regulators can inhibit the process of HDL formation. Apolipoproteins such as Apo A1 and Apo A2 play an important role in cholesterol transport, which functions to remove cholesterol from peripheral tissues to the liver. In Sentul chickens, this process is essential to support liver health, which is the main organ of lipid metabolism and toxic substance removal.

Apolipoprotein B (Apo B) is a major component of LDL (Low-Density Lipoprotein) and VLDL (Very Low-Density Lipoprotein) lipoproteins, which are involved in the transport of lipids from the liver to peripheral tissues. The increase in ApoC in T5 may indicate an increase in lipase enzyme activity that aids in the breakdown of triglycerides in lipoproteins. Lipoprotein profiles showed that HDL and LDL concentrations varied significantly between treatments (p<0.05). The HDL, known as good cholesterol, had the highest value in T3 (1.33 mg/dL) and the lowest in T5 (1.21 mg/dL). In contrast, LDL, known as bad cholesterol, showed different fluctuation patterns. Normal LDL levels in laying hens are LDL TG/5. At T1, the low LDL level was below the normal range. The function of LDL (Low-Density Lipoprotein) in the blood of laying hens is as the main transporter of cholesterol from the liver to the body tissues that need it, including the oocytes in the ovaries for yolk formation. The LDL carries cholesterol needed for cell growth and development as well as hormone production and plays an important role in the synthesis of yolk cholesterol, which accounts for about 60% of total yolk cholesterol[10-12].

Table 1: Testing regulator concentration and lipid transport in Sentul chicken layer phase

	Lipid transport regulators						
Treatment	PPAR-γ (µg/dL)	Apo-A1 (mg/dL)	Apo-A2 (mg/dL)	Apo-B (mg/dL)	Apo-C (mg/dL)	HDL (mg/dL)	LDL (mg/dL)
T1	2.11	3.97^{ab}	0.76^{ab}	0.75^{d}	0.93^{a}	2.16^{a}	0.60^{e}
T2	2.06	3.90^{a}	0.76^{a}	0.69^{cd}	0.98^{ab}	2.01^{d}	0.86^{d}
T3	2.03	3.01^{bc}	0.84^{c}	0.60^{bc}	1.05^{cd}	1.33^{bc}	1.78^{bc}
T4	2.13	2.75^{bcd}	0.88^{d}	0.53^{ab}	1.05^{c}	1.31^{b}	1.59^{b}
T5	2.07	2.76^{bcd}	1.02^{e}	0.46^{a}	1.13^{e}	1.21^{e}	2.19^{a}

[a,b,c,d]Different notations on the same line indicate real differences (p<0.05), PPAR-γ: Peroxisome proliferator-activated receptor γ, Apo-A1: Apolipoprotein A I, Apo-A2: Apolipoprotein A II, Apo-B: Apolipoprotein B, Apo-C: Apolipoprotein C, HDL: High density lipoprotein and LDL: Low density lipoprotein

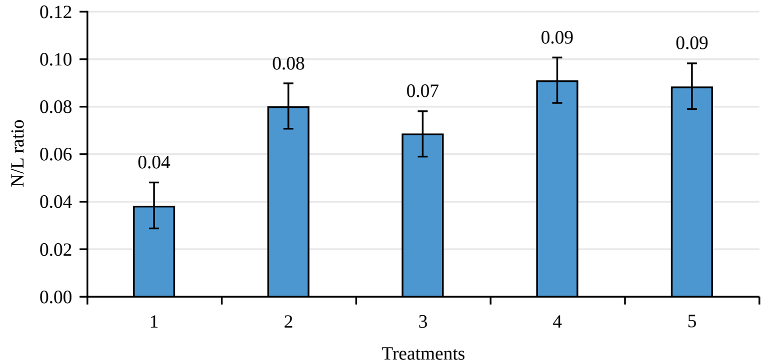

Fig. 1: Average of neutrophil-lymphocyte ratio

Table 2: Inflammation testing in Sentul chickens in the layer phase

	Treatment				
Parameters	T1	T2	T3	T4	T5
Leukocyte (10^3 cells/mm^3)	16.477^a	15.02^c	13.458^b	13.959^{bc}	13.136^b
Neutrophil (10^2 cell/mm^3)	0.376^a	0.616^{bc}	0.596^b	0.673^{cd}	0.667^{cd}
Lymphocyte (10^2 cells/mm^3)	9.755^a	7.67^b	8.677^c	7.383^c	7.517^c
IL-1β	1.39^a	1.50^a	2.1^b	3.72^c	3.80^c
IL-10	0.56^a	1.51^b	1.61^b	3.23^c	3.82^c
TNF-α (μ/L)	0.74^a	0.1^b	0.11^b	-0.77^c	-0.78^c
g-GT	0.80^a	1.35^b	1.49^b	2.66^c	2.71^c
Alkaline phosphate (U/L)	4.98^a	4.16^b	3.25^c	3.11^c	2.54^c

^{a,b}Different notations on the same line indicate real differences ($p<0.05$)

The main component of HDL that regulates cholesterol levels and allows the movement of HDL in and out of the bloodstream to collect excess cholesterol and reduce VLDL synthesis. The HDL cholesterol will transport unused cholesterol along with fat in the form of bile salts and neutral steroid hormones, resulting in low blood cholesterol, which causes a decrease in cholesterol in eggs because there is no excess cholesterol accumulated in the tissues[13,14]. A decrease in LDL may also reflect a reduced risk of cholesterol accumulation in tissues, which is often associated with impaired liver metabolism. Sentul chickens, with their adaptive immune system, require a balanced lipid profile to maintain the health of vital organs such as the liver.

Inflammation in the Sentul chicken layer phase: The results of this study showed significant changes in the values of the parameters tested, along with the administration of different treatments. The results showed T1 (control) had the highest fd leukocyte count (16.477), indicating a natural inflammatory response. Leukocytes in the normal range are 12,000-30,000 cells/μL. Based on Table 2, the average leukocyte levels in this study were 13.136×10^3/mm^3-16.477×10^3/mm^3. These levels are in the range of normal leukocyte levels. When viewed from the parameters that are closely related to inflammatory indicators are the levels of IL-1β, IL-10 and TNFα (μ/L). The results showed that T1 had the highest lymphocyte value (9.755^a), while T5 decreased significantly (7.517^c) shown in Table 2.

The highest average ratio N/L was in T5, which was 0.09 and the lowest was in T5, which was 0.04 shown in Fig. 1. Leukocytes are white blood cells that act as the body's main defense against infection or inflammation. The T5 treatment showed a significant decrease (13.136), indicating that the treatment was able to suppress the inflammatory response. The decrease in leukocytes in the treatment groups (T2-T5) indicates that the intervention is effective in reducing inflammation. Active substances such as flavonoids can act as antioxidants that can improve the number of leukocytes in the blood so as to increase the immunity of livestock[15,16]. Flavonoids act as antioxidants by preventing free radicals, directly capturing free radicals, or indirectly increasing antioxidant enzymes[17,18].

Lymphocytes play a role in the body's cellular immunity. The decrease in lymphocytes at T5 indicates that inflammatory conditions are starting to decrease, because an increase in lymphocytes usually indicates an immune response to infection. In previous research, also said that a high percentage of active lymphocyte cells will effectively prevent infection in poultry, which will significantly improve animal health[19]. Inflammatory indicators are within the normal range;

there are several indicators that have improved, including Leukocytes, Lymphocytes, IL-1β, IL-10, TNFα (μ/L) and γ-GT. The results of this γ-GT study showed a significant difference from the results of statistical analysis, indicating an improvement in liver function due to treatment.

The γ-GT levels increased after treatment and were still within normal levels. Gamma-glutamyl transferase (GGT), also known as gamma-glutamyl transpeptidase, is an enzyme that plays a crucial role in the metabolism of glutathione, an important antioxidant molecule. Its primary function is to cleave and transfer the gamma-glutamyl moiety from glutathione to amino acids or peptides, which is essential for maintaining cellular glutathione levels and protecting cells against oxidative damage caused by reactive oxygen species like hydrogen peroxide[20,21]. These inflammatory indicators are commonly used to assess liver function, as chronic inflammation can lead to liver damage characterized by an increase in these enzymes. These enzymes are known as sensitive biomarkers for liver damage[22].

Leaf extracts can be beneficial to use as an effective feed supplement in poultry to improve feed efficiency in poultry. In previous research, it was stated that Moringa leaf powder in feed resulted in significantly lower levels of egg yolk cholesterol, which could increase egg production, egg mass, feed efficiency, egg yolk color, shell thickness, β-carotene, Mg and Ca content in egg yolk, but reduced egg yolk cholesterol levels in laying hens[23-25]. The main mode of action of its active ingredients is to inhibit microbial pathogens and endotoxins in the gut and increase pancreatic activity, resulting in better metabolism and nutrient utilization[26,27]. Moringa oleifera extract was found to be more effective in controlling the tested gram-negative bacteria than gram-positive bacteria[28]. The antimicrobial activity of plants is mainly due to the presence of secondary metabolites. Plants are rich in various secondary metabolites, such as tannins, terpenoids, alkaloids and flavonoids, which *in vitro* have been found to have antimicrobial properties[29]. These active compounds in the poultry digestive tract will be able to help absorb nutrients. With improved livestock health, it is expected that the resulting production will also be better. The results of previous research also mentioned that the addition of noni fruit extract can improve nutrient digestibility and growth performance of Sentul chickens.

CONCLUSION

Noni fruit extract microcapsules (NFEM) had a positive effect on lipid profile, blood biochemistry and inflammation in Sentul chickens in the layer phase. Increasing the dose of NFEM was able to reduce LDL levels and increase HDL levels, indicating an improvement in lipid metabolism. Decreased leukocytes and lymphocytes and improved liver enzymes, showed the ability of noni extract to suppress inflammation and improve liver function. Overall, NFEM supplementation can be used as an alternative natural feed additive to improve the health and productivity of Sentul chickens.

SIGNIFICANCE STATEMENT

This study identified the beneficial effects of noni fruit extract microcapsules (NFEM) on lipid metabolism, inflammatory response and liver function in laying hens, which could be beneficial for improving poultry health and productivity through natural feed additives. This study will assist researchers in uncovering critical areas of functional phytogenic feed supplementation that have remained unexplored by many. Consequently, a new theory on the role of bioactive plant compounds in enhancing poultry physiology and performance may be developed.

ACKNOWLEDGMENT

This research work was carried out with the support of Academic Leadership Grant Universitas Padjadjaran.

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