

The Effect of Moringa Multi-Nutrient Block Supplementation on the Blood Biochemical Profile of Etawah Crossbred Goats Exhibiting Delayed Estrus and Repeated Mating

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ABSTRACT

This study investigated the impact of Moringa Multi-Nutrient Block (MMNB) supplementation on the blood biochemical profiles of Etawah crossbred (EC) goats experiencing delayed estrus and repeated mating. Fifty EC goats at the Pelaihari Breeding Centre were divided into three groups based on reproductive status: does with normal estrus cycles, doeling goats with delayed estrus, and repeat-breeder does. Each group was further subdivided to receive either MMNB supplementation or a commercial mineral block (Calphos Block, Royal®) as a control. Blood samples were collected before and after eight weeks of supplementation to assess blood urea nitrogen (BUN), glucose, and cholesterol levels. The results showed that BUN, glucose, and cholesterol values remained within normal ranges for goats, with no significant differences among groups. However, the MMNB-supplemented groups tended to have improved blood biochemical profiles compared to controls, likely due to the more balanced nutritional content of the MMNB, which includes protein, fat, minerals, and vitamins sourced from *Moringa oleifera* and other ingredients. These findings suggest that MMNB supplementation can help maintain optimal metabolic status and may support reproductive health in EC goats experiencing reproductive disorders. This study provides practical insights for goat breeders on the potential benefits of natural multi-nutrient supplementation to enhance animal health and productivity.

KEYWORDS: Blood biochemical profile, etawah Crossbreed, delayed estrus, repeated mating, moringa, multinutrient block

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I. INTRODUCTION

The Etawah crossbreed (EC) goats are a popular breed in Indonesia due to their high efficiency in producing both milk and meat [1]. Nonetheless, EC goat farmers frequently encounter challenges such as delayed estrus and repeated mating. Delayed estrus refers to female goats that have entered the estrus phase but show signs of heat later than usual, leading to postponed mating. Typically, goats reach puberty between 6 and 8 months of age. The average estrus cycle lasts about 21 days, with standing estrus averaging 36 hours, though it can vary from 24 to 48 hours depending on factors like age, breed, season, and the presence of a male [2]. Repeat mating occurs when a female goat with a normal estrus cycle has been mated two or more times with fertile males or inseminated with fertile semen but has not

pregnancy [3]. Delays in estrus can reduce reproductive efficiency [4], which negatively affects productivity [5] and farm profitability [6].

Feed plays a crucial role in influencing the health and reproductive performance of livestock. The Goats need a proper balance of energy, protein, vitamins, minerals, and access to clean water. A deficiency of protein in the rations can lead to weak estrus signs, halted estrus cycles, and repeated mating occurrences. Deficiency of essential minerals such as selenium, cobalt, copper, zinc, and iodine may cause a 5-20% reduction in growth rates and lower reproductive efficiency [4]. Incorporating natural supplements like Moringa leaves (*Moringa oleifera*) into feed has demonstrated potential in enhancing feed quality and improving animal health [7].

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Moringa leaves is known to be rich in nutrients, including protein [8, 9], carbohydrates [9, 10], fat [9, 11], vitamins [9, 12], minerals [9, 13], and antioxidants [9], all of which support vital biochemical processes in goats [14].

The feed technology multivitamin block supplement has long been developed and proven to increase the productivity and reproductivity of ruminant livestock, especially goats [15, 16, 17]. The feed supplement technology, molasses multivitamin block based on tree legumes, has been developed for sheep. The feed supplement containing *Moringa oleifera* obtained better performance and did not cause health problems in sheep [18]. The use of Moringa in the form of *Moringa Multi Nutrient Block* (MMNB) supplement feed can provide a more balanced nutritional intake and improve the blood biochemical profile, which plays an important role in the reproduction process [19].

The feed consumed and digested by livestock is absorbed and transported via the bloodstream to various organs that require nutrients. It is important to analyse blood concentrations because blood is a circulating fluid that flows throughout the body via vessels. The health status of EC goats can be assessed by examining the balance of metabolites in their blood, which is indicated by levels of blood urea nitrogen (BUN), blood glucose, and blood cholesterol. BUN is an end product of protein metabolism in the animal and is excreted through urine. It originates from ammonia in the rumen and the breakdown of amino acids. Glucose serves as the primary energy source for animal cells and is supplied both from dietary carbohydrates and internally through processes like gluconeogenesis and glycogenolysis in the liver. The absorption of glucose from the gastrointestinal tract into the bloodstream is regulated by neural signals, enterohormones (incretins), as well as the composition of the diet and intestinal microbiota. Glucose homeostasis represents the balance between glucose supply and its utilisation [12]. In ruminants, blood glucose is derived not only from dietary sugars but also from volatile fatty acids (VFA) produced during the fermentation of crude fibre. Rumen microbes ferment carbohydrates into VFA, primarily acetate, propionate, and butyrate, which serve as the main energy sources for ruminant animals. The lipid content in the blood of both ruminant and monogastric animals is influenced by the type and amount of feed consumed [20]. Blood lipid profiles are important indicators used to assess the health and productivity of livestock.

Today, measuring the blood urea nitrogen (BUN) levels in ruminant animals is a widely used method to assess their protein status, both in livestock production studies and for maintaining animal health [21]. BUN serves as an important indicator of nitrogen metabolism within the rumen and can be utilised as a measure of nitrogen balance in cattle and sheep [22]. Blood glucose testing is conducted because research has shown that ruminants require glucose throughout all stages of life, with their glucose needs mirroring the pattern of protein requirements [23]. Cholesterol is a key plasma lipid essential for the body, as it

forms a structural part of cell membranes and acts as a precursor for the synthesis of bile acids and steroid hormones [24].

This study aims to compare the blood biochemical profiles of EC goats experiencing delayed estrus with those undergoing repeated mating, both before and after supplementation with *Moringa Multi Nutrient Block* feed. By analysing the changes in these blood biochemical profile, the study seeks to identify effective strategies to enhance the reproductive health of EC goats and offer practical recommendations for breeders in managing their animals' nutrition and overall health.

II. MATERIAL AND METHODS

A. Experimental Goats

This study used 50 etawah crossbreed (EC) goats at the Pelaihari Breeding Centre.

B. Experimental Feed Ingredients

The feed ingredients used were *Moringa Molasses Multivitamin Block* (MMNB) supplement feed as the treatment feed, and commercial mineral block feed Calphos Block, Royal® made in Turkey, and commercial concentrate feed Nuvo NC62® produced by PT. Charoen Pokphand Indonesia TBK, which is currently used by the Pelaihari Breeding Centre as the control feed. The MMNB supplement feed formulation is presented in Table 1, while the nutritional content of the MMNB supplement feed, commercial mineral block, is presented in Table 2, and the nutritional content of the concentrate feed is presented in Table 3.

Table 1. The composition of MMNB supplement feed ingredients.

Feed Ingredients	Jumlah (%)
<i>Moringa oleifera</i> leaf flour	10
Palm oil cake	13
Fish meal	10
Fine rice bran	10
Molasses	25
Lime	5
Salt	10
Cement	10
Mineral mix	2
Quantity	100

Table 2. Nutritional content of MMNB and Calphos Block, Royal® supplement feed

Nutrient Content	MMNB ¹	Calphos Block, Royal® ²
Dry matter	86.67%	97.69%
Moisture Content	13.33%	2.31%
Ash	33.42%	42.61%
Crude Protein	15.36%	5.45%
Crude Fiber	7.31%	3.78%
Fat ¹	3.71%	2.45%
Ca	-	1.22%

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P	-	0.11%
Na	-	37.79%
Mg	-	1.15%
Mn	-	1,550 mg
Fe	-	770 mg
Zn	-	650.10 mg
Cu	-	500 mg
I	-	60.15 mg
Co	-	40 mg
Se	-	20.65
Vitamin A	-	10,000,000 IU
Vitamin D3	-	2,000 IU
Vitamin E	-	20 mg

Note: ¹ Results of analysis of Animal Nutrition and Feed Laboratory, Animal Science Department, Faculty of Agriculture, University of Lambung Mangkurat, Banjarbaru – Indonesia.

² According to the packaging label.

Table 3. Nutritional content of concentrated feed.

Nutrient Content	Quantity
Dry matter	Max 14%
Ash	Max 12%
Crude Protein	Min 14%
Crude Fat	Max 7%
Calcium (Ca)	0.6 – 1.2%
Phosfor (P)	0.8%
Total Aflatoxin	Max 200 ppb
Neutral Ditergen Fibre (NDF)	Max 35%
Total Digestible Nutrient (TDN)	Min 68%

Source: PT. Charoen Pokphand Indonesia TBK

C. Experimental Design.

This study involved 50 EC goats, categorised into three distinct groups based on reproductive status and treatment allocation:

Group 1: Consisted of 10 doe goats with a normal estrus cycle and a history of giving birth.

Group 2: Included 20 virgin goats (doelings) that had not exhibited estrus but were expected to do so and be mated. These were split into:

- 10 goats receiving the MMNB supplement as the treatment group.
- 10 goats receiving a Calphos Block, Royal® as the control group.

Group 3: Comprised 20 doe goats that had been mated more than twice without becoming pregnant. These were also divided into:

- 10 goats were given the MMNB supplement as the treatment group.
- 10 goats were provided with the Calphos Block, Royal® as the control group

Each group was established to enable comparison of blood biochemical profiles and reproductive responses between goats with different reproductive challenges and supplement regimens.

The use of both treatment and control groups within Groups 2 and 3 allows for the evaluation of the specific effects of the MMNB supplement relative to standard mineral supplementation, following principles of controlled experimental design and replication as recommended in livestock nutrition studies.

D. Blood Sampling

Blood samples were collected twice: initially at the first week before the administration of the MMNB supplement, and again after eight weeks of supplementation. Sampling was performed in the morning, before feeding, by drawing blood from the jugular vein using a venoject. This approach aligns with standard practices, where the jugular vein is commonly used for blood collection in goats due to its accessibility and ease of handling. After collection, the blood samples were centrifuged at 2,000 rpm for 30 minutes to separate plasma and serum. The plasma was then stored at -18°C in a freezer until all samples were ready for analysis. Subsequently, the samples were transported to the South Kalimantan Provincial Health Laboratory in Banjarmasin for the measurement of blood urea nitrogen (BUN), glucose, and cholesterol levels.

E. Data Analysis

The biochemical blood profile data from goats with normal cycles, delayed estrus, and repeated mating were analysed using ANOVA in a Completely Randomised Design. Comparisons between subgroups of goats fed the MMNB supplement and those receiving the Calphos Block, Royal® were conducted using an *independent sample t-test*. Additionally, *paired sample t-tests* were used to compare blood profiles before and after supplementation within each group. All statistical analyses followed the procedures outlined by Steel and Torrie (1993) and were performed using SPSS® IBM® Version 26.

III. RESULTS AND DISCUSSION

A. Comparison of Blood Biochemical Profiles of Goats with Normal Cycles, Late Estrus, and Repeated Mating

The average levels of blood urea nitrogen (BUN), glucose, and cholesterol in the blood of experimental goats with normal cycles, late estrus, and repeated mating are presented in Table 4.

Table 4. Comparison of blood biochemical profiles of experimental goats.

Treatment	BUN (mg/dL) (Rata-rata ± SEM)	Glucose (mg/dL) (Rata-rata ± SEM)	Cholesterol (mg/dL) (Rata-rata ± SEM)
Normal Cycle	14.42±0.60	73.44±2.88 ^{ab}	106.67±4.60
Late in heat	17.58±3.00	70.17±2.78 ^b	73.44±2.88
Repeated Mating	14.63±0.66	62.48±1.67 ^a	94.24±5.16
References	6 – 27 ¹	40 – 60 ²	43 – 103 ¹

Notes: ^{a,b} Mean with different superscripts on the same column showed a significant difference (p<0.05)

¹. [32]

². [30]

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Blood Urea Nitrogen (BUN)

Table 4 shows that BUN levels among normally cycling does, maiden goats with delayed estrus, and repeat-breeder does did not differ significantly ($p>0.05$), with all values remaining within the normal range for goats [25]. These normal BUN values indicate proper protein metabolism in PE goats, and similar BUN levels have also been found in male goats [26, 27], suggesting that sex does not affect blood urea levels [25]. The BUN standards applied are general and do not account for physiological status or sex. However, there is a tendency for higher BUN levels in maiden goats with delayed estrus and repeat-breeder does compared to normally cycling does, still within the normal range as reported by [28], though further research with larger sample sizes is needed.

Normal BUN levels indicate adequate, or possibly excessive, dietary protein intake, as urea production is directly related to protein consumption. Excess protein can negatively impact fertility by causing metabolic disturbances, hormonal imbalance, and a uterine environment less conducive to embryo development. In cattle, excess dietary protein or urea is also linked to reduced conception rates and reproductive disorders, especially when energy balance is not maintained [29]. To confirm these effects, further studies with stricter nutritional control and larger animal numbers are required.

Glucose

Blood glucose levels in normally cycling does, doeling goats with delayed estrus, and repeat-breeder does showed no significant differences ($p>0.05$), with all values remaining within the normal range for ruminants [23]. However, blood glucose levels in doeling goats with delayed estrus were significantly higher than in repeat-breeder does ($p<0.05$), while repeat-breeder does tended to have lower levels than normally cycling does, though not statistically significant. These normal blood glucose levels indicate proper carbohydrate metabolism in PE goats [30].

Previous studies on male PE goats also reported normal blood glucose levels [26, 27], though lower than those in normally cycling does, suggesting potential influences of sex and age on glucose levels. The lower glucose levels in repeat-breeder does are suspected to result from dietary energy deficiency, which may disrupt reproductive hormone production, leading to estrus cycle irregularities and repeat breeding. In ruminants, blood glucose levels are influenced by carbohydrate intake and metabolism, as well as gluconeogenesis and glycogenolysis processes that maintain stable blood glucose levels [31].

Cholesterol

Table 9 shows that blood cholesterol levels in cycling, doeling goats with delayed estrus, and repeat-breeder goats did not differ significantly ($p>0.05$), and all values were within the normal range of 43–103 mg/dL [32]. This indicates normal fat metabolism, particularly cholesterol, in EC goats.

Previous studies on male PE goats reported normal cholesterol levels (61.38 ± 4.85 mg/dL and 77.00 ± 9.04 mg/dL)

[26, 27], which were lower than those found in female goats in this study, suggesting females may have higher cholesterol levels, although further research with larger samples is needed. The study also found that EC does with repeated mating and does with delayed estrus have lower cholesterol levels than does with normal cycles. According to [33], low cholesterol can cause silent or short estrus, prolong anestrus, reduce pregnancy rates, and trigger postpartum anestrus. This may explain delayed or absent estrus in these goats. These findings align with research on dairy cows showing lower cholesterol in repeat breeders compared to fertile cows. Cholesterol, produced by the liver, is essential for steroid hormone synthesis, vitamin D, and bile acids. Low cholesterol, possibly due to insufficient dietary fat, can impair reproduction by suppressing estrus and ovulation, leading to silent heat, prolonged anestrus, or repeated mating.

B. Blood Biochemical Profile Before and After Supplementation

Blood Urea Nitrogen

The average levels of Blood Urea Nitrogen (BUN) of experimental goats with late in heat, and repeated mating before and after being given feed supplement for eight weeks, are presented in Table 5.

The supplementation of MMNB and Calphos Block, Royal® in doeling goats with delayed estrus significantly increased ($p<0.05$) BUN levels. This increase is due to the complete nutrient content, especially protein and minerals, which improves protein metabolism in EC goats. The MMNB supplement, containing higher protein (15.36%), caused BUN levels to exceed the normal range, while Calphos Block, Royal®, higher in minerals but lower in protein (5.45%), increased BUN within normal limits according to [32]. BUN levels in the treatment group were higher than in the control group (17.57 vs. 11.56 mg/dL), influenced by protein and minerals from ingredients such as moringa leaves and fish meal in MMNB. Excess protein negatively affects fertility by disrupting energy efficiency and the secretion of gonadotropin and progesterone hormones, which are essential in the estrus cycle. Overfeeding protein lowers uterine pH during the luteal phase, disturbing the uterine environment and estrogen secretion that triggers estrus, thus causing delayed estrus in doeling goats.

Table 5. Levels of BUN (mg/dL) in the experimental goats with late in heat, and repeated mating before and after being given feed supplement.

Observation	Late in heat		Repeated Mating	
	Treatment	Control	Treatment	Control
Before	13.50±	15.59±	13.15±	16.27±
	0.72 ^a	1.12 ^a	0.83 ^a	0.70 ^a
	31.07±	27.15±	30.78±	30.67±
After	1.32 ^b	1.76 ^b	1.90 ^b	0.98 ^b
References	6 – 27 ¹			

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Notes:

^{a,b} Mean with different superscripts on the same column showed a significant difference ($p < 0.05$)

¹. [32].

Supplementation with MMNB and Calphos Block, Royal® in does experiencing repeat breeding significantly increased ($p < 0.05$) BUN levels, with a greater increase compared to the control group (17.63 vs. 14.40 mg/dL), exceeding the normal range according to [32]. The higher BUN levels in the treatment group were mainly due to the higher protein content in the MMNB supplement. Excess urea in the blood can reduce pregnancy rates because elevated ammonia or urea levels disrupt oocyte maturation and are toxic to the ovum [29], leading to fertilisation failure and repeat breeding.

Glucose

The average levels of glucose of experimental goats with late in heat, and repeated mating before and after being given feed supplement for eight weeks, are presented in Table 6.

Table 6 indicates that the blood glucose levels of doeling goats in the treatment group rose after receiving the MMNB supplement, although this increase was not statistically significant ($p > 0.05$). Conversely, the control group showed a decrease in blood glucose levels following supplementation with Calphos Block, Royal®, but this reduction was also not significant. The rise in glucose levels observed in the treatment group can be attributed to the more comprehensive and higher nutritional value of the MMNB. The MMNB contains minerals and nutrients derived from various feed ingredients listed in Table 3, five of which serve as carbohydrate (glucose) sources as well as mineral sources, including moringa leaf flour, palm kernel meal, fish meal, fine rice bran, and molasses.

Table 6. Levels of glucose (mg/dL) in the experimental goats with late in heat, and repeated mating before and after being given feed supplement.

Observation	Late in heat		Repeated Mating	
	Treat-Ment	Control	Treat-ment	Control
Before	70.83±	69.45±	63.64±	61.18±
	2.90	5.03	3.02	1.10
After	63.33±	67.73±	59.36±	60.18±
	1.73	2.23	2.11	1.63
References	40 – 60 ¹			

Notes:

^{a,b} Mean with different superscripts on the same column showed a significant difference ($p < 0.05$)

¹. [30]

According to Table 6, blood glucose concentrations in goats from repeated mating declined in both the treatment and control groups following supplementation with MMNB and Calphos

Block, Royal®. Nevertheless, this reduction was not statistically significant ($p > 0.05$). These findings imply that supplementation may contribute to maintaining blood glucose levels within the normal range for goats and sheep, as noted by [30]. The observed decrease is likely attributable to the supplements more comprehensive and higher-quality nutritional composition. A balanced and nutrient-dense diet enhances metabolic efficiency and facilitates greater glucose uptake from the bloodstream.

Cholesterol

The average levels of cholesterol of experimental goats with late in heat, and repeated mating before and after being given feed supplement for eight weeks, are presented in Table 7.

Table 7. Levels of cholesterol (mg/dL) in the experimental goats with late in heat, and repeated mating before and after being given feed supplement.

Observation	Late in heat		Repeated Mating	
	Treat-ment	Control	Treat-ment	Control
Before	110.73±	106.18±	84.91±	104.45±
	6.18a	6.82	6.33	6.58
After	128.82±	118.45±	99.00±	112.36±
	4.79 ^b	6.37	5.27	6.71
References	43 – 103 ¹			

Notes:

^{a,b} Mean with different superscripts on the same column showed a significant difference ($p < 0.05$)

¹. [32].

The cholesterol levels observed in the experimental goats reflect the impact of supplementary feed composition on lipid metabolism. According to [32], normal blood cholesterol levels for goats and sheep range between 43–103 mg/dL. In this study, the treatment group (given MMNB supplement), cholesterol levels remained within the normal range (43–103 mg/dL) both before and after supplementation. The 18.09 mg/dL increase post-supplementation correlates with MMNB's higher fat content (3.71%) and mineral profile, which may enhance lipid metabolism [34]. The control group (given Calphos Block, Royal®), cholesterol levels exceeded the normal range post-supplementation, rising by 12.23 mg/dL. This suggests that Calphos Block, Royal® mineral-focused formulation (lower fat: 2.45%) had a less pronounced effect on lipid pathways compared to MMNB [34].

Table 7 demonstrates that the cholesterol concentrations in the treatment group of experimental goats, both before and after receiving supplemental feed, remained within the normal reference range for goats and sheep, which is 43–103 mg/dL as defined by [32]. In contrast, the control group exhibited cholesterol levels above this normal range before and after supplementation. Additionally, PE with regular estrus cycles had an average blood cholesterol level of 106.67 ± 4.60 mg/dL, slightly higher than the established normal range. This pattern

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was also observed in the group of doeling goats, indicating that goats with normal estrus cycles may have cholesterol levels at or just above the upper limit of the typical range for goats and sheep [35].

CONCLUSION

The results of this study demonstrate that supplementation with Moringa Multi-Nutrient Block (MMNB) in Etawah crossbred goats experiencing delayed estrus and repeated mating maintained blood urea nitrogen (BUN), glucose, and cholesterol levels within normal physiological ranges for goats. Although there were no statistically significant differences between the MMNB and control groups, goats receiving MMNB tended to show improved blood biochemical profiles compared to those receiving standard mineral supplementation. The balanced nutritional content of MMNB, which includes higher levels of protein, fat, and minerals, likely contributed to a better metabolic status in the supplemented goats. These findings suggest that MMNB is a safe and effective dietary supplement that can help support metabolic health and potentially enhance reproductive performance in EC goats facing reproductive disorders. Further research with larger sample sizes and longer supplementation periods is recommended to confirm these benefits and to explore the effects of MMNB on reproductive outcomes in greater detail.

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