Reproductive efficiency and serum progesterone concentration on dairy cattle based on blood urea nitrogen (BUN) concentrations

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Abstract

The objective of this study was to identify the levels of Blood Urea Nitrogen (BUN) and its relation to the reproductive efficiency and serum progesterone concentration in lactating Holstein Friesian dairy cattle. Data were obtained by stratified random sampling from population to get 18 dairy cows, which divided into three groups. The first, second, and third groups each with milk yield of less than 17, between 17-21, and over 21 liters/day, and with calving interval of less than 365, between 365-450 and more than 450 days. Blood was collected from the coccygeal vein at the time of insemination (D0), 7 day (D+7) and 22 days (D+22) after insemination. BUN and progesterone concentration were measured. Data analysis showed no significant difference (P>0.05) on the observed parameters. Data regrouping based on BUN concentration showed that the mean of services per conception (S/C) was lower and conception rate (CR) was higher (P<0.05) in dairy cattle having BUN<18 than those of with BUN>18 mg/dL. The mean of progesterone concentration at D+22 was higher (P<0.05) than those at D0 and D+7 in pregnant dairy cows and with BUN<18 mg/dL, as well as those at D+22 in non-pregnant dairy cows with BUN>18 and BUN<18 mg/dL. This study concluded that high BUN concentration affect the reproductive efficiency of dairy cows by decreasing CR and increasing progesterone serum concentration.

Keywords: Reproductive efficiency, progesterone, BUN, milk yield
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الكفاءة التناسلية وتركيز البروجسترون في مصل دم الأبقار الحلوبة بالاعتماد على تراكيز نيتروجين اليوريا في الدم

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الخلاصة

هدفت هذه الدراسة إلى التعرف على مستويات نيتروجين اليوريا في الدم وعلاقته بالكفاءة التناسلية وتركيز البروجسترون في المصل في الأبقار الحلوبية هولشتاين فريزيان. تم الحصول على البيانات عن طريق أخذ عينات عشوائية طبقية من ١٨ بقرة حلوب، والتي تنقسم إلى ثلاث مجموعات. المجموعات الأولى والثانية والثالثة مع كل محصول من الحليب أقل من ١٧، بين ١٧-٢١ ، وأكثر من ٢١ لترا / يوم، ومع فترة بين الولادات أقل من ٣٦٥، بين ٣٦٥-٤٥٠ وأكثر من ٤٥٠ يوما. تم جمع الدم من الوريد العصعصي في وقت التلقيح (اليوم صفر) و ٧ أيام و ٢٢ يوما بعد التلقيح. تم قياس نيتروجين اليوريا في الدم وتراكيز البروجسترون. أظهر تحليل البيانات عدم وجود فرق معنوي (P> 0.05) في النتائج. أظهرت إعادة تجميع المعطيات على أساس تركيز نيتروجين اليوريا في الدم أن عدد مرات التلقيح لكل حمل كان أقل وان معدل الاخصاب كان أعلى (P <0.05) في الأبقار الحلوب بعد نيتروجين اليوريا في الدم أقل من ١٨ ملغم / ديسيلتر. كان متوسط تركيز البروجسترون في اليوم ٢٢ بعد التلقيح اعلى (P <0.05) من التركيز يوم التلقيح و ٧ ايام بعد التلقيح في أبقار الحليب الحوامل التي لها تركيز نيتروجين اليوريا في الدم اقل من ١٨ ملغم/ ديسيلتر، وكذلك الحال في اليوم ٢٢ بعد التلقيح للابقار غير الحوامل

التي كان تركيز نيتروجين اليوريا في الدم اقل او اكثر من ١٨ ملغم/ ديسيلتر. يستنتج من هذه الدراسة أن تركيز نيتروجين اليوريا في الدم العالي يؤثر على الكفاءة التناسلية للأبقار الحلوب عن طريق تقليل معدل الاخصاب وزيادة تركيز البروجسترون.

Introduction

Nutrition management plays an important role in dairy farming. Feeding with high protein content can stimulate high milk production (1). Dairy farmers generally increase the amount of feeding in order increasing milk production, but increasing the amount of feed protein was known to be detrimental to reproductive performance (2,3). The final product of protein metabolism in ruminants is urea which then circulates in the bloodstream (3). There is a positive correlation between BUN and MUN and their concentrations in cattle body tend to be balanced (5,6). Therefore, both MUN and BUN measurements could be utilized as important information for farmers about the nutritional status and health of cattle (3). High BUN concentration indicated an inefficient usage of nitrogen intake for both growth and milk production (7). High feed proteins in dairy cows was reported to be associated with decreased conception rates (8), uterine pH (9,10), ovarian function and increased services per conception (11). High blood urea delayed the clearance of uterine contamination and decrease immunity, the secretion of K, Mg, and P in lactating cows and caused hormonal imbalances (12,13). High concentrations of urea nitrogen in dairy cows causes a negative effect on reproduction because it inhibited follicular development and fertilization, decreased the binding of luteinizing hormone to its receptors on ovaries as well as progesterone binding capacity to ovaries receptor (14).

The aim of this study was to identify the profile of progesterone and Blood Urea Nitrogen (BUN) concentrations and their relation to reproductive efficiency in dairy cows.

Materials and methods

This study was conducted on dairy cows with a total population about 1000 in small stakeholder farms at Precet Sumbersuko, Wagir, Malang Regency during the period from May 2017 to September 2017. The criteria of dairy cows being studied were non pregnant and in the lactation phase.

Data collection procedures

The data were collected from 100 non pregnant, lactating dairy cows based on age, parity, BCS (1-9), milk yield and reproduction based on farmers' and inseminator's recording data. On the basis of 50 cows were randomly isolated to conduct a survey on reproductive efficiency and feed given (grass and concentrate). From 50 cows that have been surveyed 18 cows with the criteria of BCS 4-7 (scale 9), parity 2 and age ±4 years were selected and grouped into three of 6 each based on the quantity of milk yield and reproductive efficiency: First group (KI): dairy cows with milk yield less than 17 liters per day, and calving interval rate less than 365 days, Second group (K2): dairy cows with milk yield 17 to 21 liters per day with Calving Interval 365 up to 450 days, third group (K3): dairy cow with milk yield more than 21 liters per day and calving interval rate more than 450 days. At estrus, cows were artificially inseminated and blood was collected from the coccygeal vein at D0 (time of AI), D+7, and D+22. The pregnancy was examined three months after insemination by rectal palpation.

Blood Urea Nitrogen (BUN) was measured by means of Berthelot method using urea nitrogen diagnostic kit (Pointe Scientific, Inc), and expressed as mg/dl while progesterone concentration was measured by ELISA test using diagnostic progesterone kit (DRG Diagnostic, Inc).

Data analysis

The analysis was conducted to identify the homogeneity of sample data with sub-population (50 with 100 dairy cows) based on parameters of age, parity, milk yield, and BCS. Similarly, the homogeneity of sample data between the 18 cows to be further explored with 50 cow samples based on S/C, CI, DO, and feed parameters. Blood Urea Nitrogen (BUN), S/C and CR values were analyzed to determine the possible differences between groups. Further analysis of milk yield, reproductive efficiency, and feeding factors were performed by regrouping based on BUN concentrations (BUN ≥ 18 and BUN<18 mg/dl). Analysis of progesterone concentrations was based on criteria for combination of BUN concentrations (BUN ≥18 and BUN <18), and pregnancy status (pregnant or non-pregnant). The overall analysis was performed by analysis of variance (ANOVA) of SPSS 20.0 for Windows program at 95% confidence level.

Results and discussion

Data obtained from 100 cows taken randomly from the population and the data of 50 cows samples have a suitability, i.e. on the average of age (4.86 and 4.19 years), milk yield (19.78 and 19.26 liters/day), parity (2.35 and 2.32), and BCS (4.92 and 4.88). Mean of BUN concentrations were not significantly different (P>0.05) among the groups (Table 1). High milk production did not affect BUN concentrations in these cattle. In dairy cows, milk production can be affected by several factors such as

breed, management, environmental conditions, feed, and lactation phase and health status condition (15). BUN is the end result of nitrogen metabolism in ruminants (7).

The parameter grouping in Table 1 was based on the difference of milk yield and CI, so each of the two parameters was significantly different (P<0.05). A rhythmic calving interval will have a positive impact on milk production (16), a calving interval of more than 400 days was known to have an effect on increased milk production (17), which was in line with increasing S/C (18). It was probably related to the concentrations of nitrogen urea in the body that also contaminate the cow's uterine fluid thus disrupting conception (13).

Table 1. Milk yield, reproductive efficiency and BUN on experimental groups

Parameters	K1	K2	K3
milk yield (kg/cow/day)	13.17±0.83[a]	19.17±0.60[b]	27.33±1.84[c]
CI (days)	357.33±1.91[a]	409.17±8.73[b]	505.33±35.61[c]
S/C	1.33±0.21[a]	3.83±0.31[b]	6.67±0.33[c]
DO (days)	66.17±7.28[a]	55.33±4.85[a]	59.50±8.13[a]
BUN (mg/dl)	14.92±1.59[a]	16.58±1.14[a]	15.33±1.52[a]
CR	33.33%	50%	33.33%

Different superscripts in one row showed a significant difference (P<0.05); G= Groups; DO= Days open; S/C = Services per conception; CI= Calving interval; BUN= Blood urea nitrogen; CR = Conception rate; K1= milk yield <17liters/day, calving interval<365 days; K2= milk yield 17-21 liters/day, calving interval 365-450 days; K3= milk yield >21 liters/day, calving interval>450 days; replicates = 6.

The data of blood urea nitrogen (BUN) and conception rate (CR) obtained on groups in Table 1 did not significantly different (P>0.05), thus then regrouped based on BUN concentrations (BUN> 18 and BUN <18 mg/dL), added with feed intake (grass, concentrate, and concentrate to grass ratio) parameters. The result showed that the mean of S/C was lower and CR was higher both significantly (P<0.05) in dairy cattle having BUN<18 than those of with BUN>18 mg/dL (Table 2).

Table 2. Milk yield, reproductive efficiency and feed based on BUN content

Parameters	BUN<18 mg/dL	BUN>18 mg/dL
BUN (mg/dL)	13.67±0.61[a]	19.50±0.52[b]
milk yield (liter/day)	18.75±2.08[a]	22.17±2.02[b]
Reproductive efficiency		
S/C	3.50±0.69[a]	4.83±0.87[b]
CR	58.33%[a]	???? 0%[b]
DO (days)	62.17±5.62[a]	56.67±3.44[a]
CI (days)	424.00±26.68[a]	423.83±21.90[a]
Feed		
Concentrate (C) kg/cow/day	10.5 ±2.95[a]	10.33 ±0.47[a]
Grass (G) kg/cow/day	33.33 ±4.24[a]	35 ±2.31[a]
C/G	32.42±3.36%[a]	29.52±0.95%[a]

Different superscripts in a row showed significant differences (P<0.05); BUN = Blood Urea Nitrogen; S/C = Services per conception; CR = Conception rate; DO = Days open; CI = Calving interval; C/G = concentrate to grass ratio.

Urea nitrogen in cow body fluids largely derives from a high protein intake of feed; the addition of concentrate in dairy feed will affect the quality and composition of milk (19). Increased feeding of high rumen degradable protein in dairy cows through the addition of concentrates potentially increases milk production by increasing protein synthesis in rumen (20). In this study the C/G values were not significantly different (P>0.05), whereas BUN was significantly different (P<0.05) (Table 2), indicating inefficient metabolism of protein in dairy cows with high BUN. High BUN concentrations indicate an inefficient utilization of nitrogen intake in the body for both growth milk productions (7). High plasma or milk urea indicates a high rumen ammonia concentrations. Excess ammonia will diffuse to the bloodstream and converted to urea in the liver (20). Feed with high protein content will result in high concentrations of urea nitrogen in plasma and milk (21). Higher concentrations of urea nitrogen in cows with lower concentrate intake indicate a lack of synchronization between energy and protein in the rumen (20). Feed consumption with a high content of rumen degradable protein in dairy cows is closely related to increased BUN concentrations (5, 22) and decreased pregnancy rates (10).

Higher milk production was accompanied with higher BUN concentrations (Table 2). BUN concentrations are one of the renal function evaluation criteria that reflect glomerular filtration rate (23). High BUN concentrations indicate an imbalance of nitrogen metabolism. High intake of protein intended to increase milk production and leaves a higher rumen undegradable protein that ends up as BUN and MUN. However, different milk yields obtained from cows fed the same quality and quantity of feed (Table 2)

can be caused by genetic factors. There are potential genetic markers and genes for the production characteristic in dairy cows, i.e. AA, AG, and GG. Cattle with AA genotype have higher milk yields than cows with GA or GG genotype (24). The genotypes AA, AG, and GG have the frequency of 0.07, 0.34, and 0.59, respectively, in the livestock population (25). In addition to genetic factors, prolactin (PRL) concentrations in the circulatory system are positively correlated with photoperiodic (26), but in this study the photoperiodic of all cows was the same.

BUN concentrations and reproductive efficiency

The reproductive efficiency of dairy cows in the tropical area is relatively low with more repeat breeder cases (27) and the average of first artificial insemination after parturition is longer (28) than dairy cows in sub-tropical regions. In addition, BUN concentrations above 18 mg/dL may decrease uterine pH and decrease fertility in dairy cows (12), by affecting the motility and viability of sperm in female reproductive tract so as to decrease pregnancy rate. The occurrence of repeat breeding in dairy cows may also be due to oocyte disorders (29).

In this study the reproductive efficiency based on CR and S/C was better ($P < 0.05$) in the BUN<18 mg/dL group compared with BUN> 18 mg/dL group, but the DO and CI were relatively equal ($P > 0.05$) (Table 2). Dairy cows that have higher BUN concentrations have higher milk production, higher S/C and lower CR ($p < 0.05$), whereas DO and CI were both equal ($P > 0.05$). This fact indicates that BUN concentrations exceeding the threshold of 18 mg/dL interfere with reproduction at the time of fertilization. The normal range of nitrogen urea concentrations in cows is 8 to 25 mg/dL (21). Several previous studies had shown that concentrations of nitrogen urea in cows greater than 18 mg/dL may cause some negative effects on reproduction, decreased pregnancy rate (12),increasing S/C, decreasing uterine pH and affecting the balance of reproductive hormone concentrations (2,14).

Many factors can affect CR in lactating cows including metabolic factors, infectious diseases and environmental conditions. Metabolism in cows with high milk production causes a lower CR value. This may be associated with lower body condition score in cows with high milk production (18). The decrease in CR value may also be associated with an increase in protein feed (30). This is in line with previous studies suggested that BUN concentrations above 19 mg/dL will decrease uterine pH and decrease the fertility of dairy cows (31). High rumen degradable protein content in cow will cause urea concentrations in blood, milk and urine to increase which causes fertility problems, decreased energy availability, environmental pollution and economic losses (32).

High concentrations of urea nitrogen in body fluids can decrease pregnancy rates (12), ammonia concentrations will increase so as to delay the clearance of uterine contamination and decrease the functioning of the immune system, decrease the secretion of K, Mg, and P in lactating cows and cause hormonal imbalance by decreasing binding luteinizing hormone to the ovary receptors (8,14). Progesterone is an essential hormone in the entire process of cow reproduction from ovulation to maintaining pregnancy, development of the mammary glands and the associated neurobehavioral role of sexual response (33).

Serum progesterone concentrations

Progesterone is produced by the corpus luteum in the ovaries controlled by the prolactin (PRL). Progesterone concentrations in the circulation are fluctuating following the estrous cycle, low in the follicular phase and high in the luteal phase. In dairy cows that heat and then inseminated and pregnant, the originally low concentration of progesterone in the blood will rise and remain high until the parturition. In cows that were not fertilized after insemination, the endometrium will produce prostaglandin (PG) F2α starting around day 14. The PGF2α serves to regress the corpus luteum, so that the corpus luteum begins to degenerate on day 17, followed by a drop in progesterone to basal concentrations on days 20 to 23 as estrus return (34). Thus the physiological concentrations of progesterone in serum differed between pregnant and non-pregnant cows. Therefore, the progesterone concentration in this study was regrouped based on BUN concentrations then divided again in pregnant and non-pregnant cows (Fig. 1).

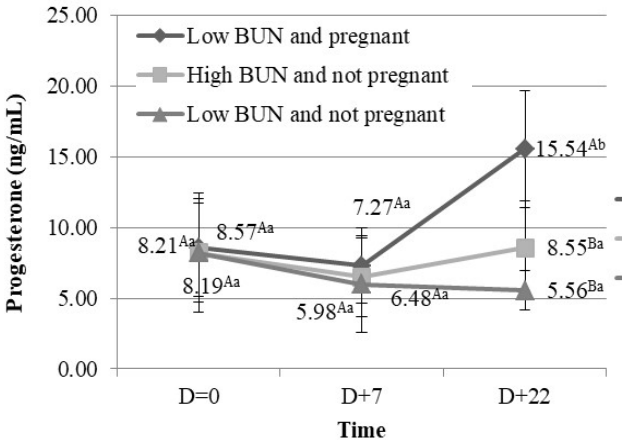

Figure 1. Serum progesterone concentration of cow group with low BUN and pregnant.

High BUN and not pregnant, also low BUN and not pregnant at AI (D0), 7th day (D+7), and 22nd (D+22) after AI. Description: The letters A and B are significantly different ($P < 0.05$) between treatments, a and b are significantly different ($P < 0.05$) between sampling days.

In this study, the progesterone D+22 in the cows that were non pregnant with high BUN or low BUN concentration were not differ significantly. There was a similar pattern on progesterone concentrations D0, D+7, and D+22 in non-pregnant cow with high BUN concentration and pregnant cow with low BUN concentration. However D+22 serum progesterone concentrations in non-pregnant cow with BUN> 18 mg/dL were lower than serum progesterone D+22 in pregnant dairy cow with BUN <18 mg/dL. This was in accordance with previous reports (1,13,14) stated that high concentrations of BUN may decrease progesterone concentration in dairy cows. Feed intake and high milk production have been associated with increased portal blood flow, and increased progesterone clearance by the liver (35). Low concentrations of progesterone at the beginning of luteal phase negatively affect the survival of the embryo (36). This could be the cause of the failure of pregnancy in cows with high BUN (Figure 1). Progesterone is produced by the corpus luteum in the ovaries that is controlled by the prolactin hormone, while the prolactin hormone as discussed previously also plays a role in milk production in response to nutrient intake. The molecular mechanisms of BUN effects on serum progesterone concentrations are unknown, therefore it needs for further researches involves measurements of hormone concentrations of PRL and associated molecules.

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