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CORRELATION BETWEEN BLOOD METABOLITE AND REPRODUCTIVE PERFORMANCE OF LACTATING HOLSTEIN FRIESIAN CROSSBRED COWS IN SMALLHOLDER FARMERS

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ABSTRACT

This study examined the correlation between the concentration of blood metabolites (triglyceride, albumin, phosphorous) on the reproductive performances (S/C and PPM) of lactating Holstein Friesian crossbred (HFC) cows. Blood samples were collected through the caudal vein 8 hours after feeding and the plasma was analyzed at the Integrated Research and Testing Laboratory of Universitas Gadjah Mada (LPPT UGM). Samples were stored at 5° C. Triglyceride, albumin, and phosphorous were measured using glucose oxidase-phenol 4-aminoantipyrine (GOD-PAP) method, photometric bromocresol green method, and cypress diagnostic, respectively. Blood metabolites data were analyzed using Pearson correlation model. The results showed that the correlation coefficients of triglycerides, albumin, and phosphorus levels on PPM and S/C were -0.521 and -0.650; -0.447 and -0.612; -0.513 and -0.700, respectively. In conclusion, there are significant negative correlation between triglyceride, albumin and phosphorus levels on the PPM, and very significant negative correlation between triglyceride, albumin and phosphorus levels on the S/C of lactating HFC cows.

Key words: blood metabolite, Holstein Friesian Crossbred, lactation cows, reproductive performance

ABSTRAK

Penelitian ini bertujuan mengetahui korelasi metabolit darah (trigliserida, albumin, dan fosfor) terhadap kinerja reproduksi sapi perah peranakan Friesian Holstein (PFH) laktasi yang meliputi service per conception (S/C) dan postpartum mating (PPM) di peternakan rakyat. Sampel darah diambil 8 jam setelah pemberian pakan melalui vena caudalis menggunakan vacum holder dengan EDTA kemudian disimpan pada suhu 5° C dan dianalisis di Laboratorium Penelitian dan Pengujian Terpadu Universitas Gadjah Mada (LPPT UGM). Data kinerja reproduksi meliputi S/C dan PPM didapatkan dari hasil recording. Parameter yang diamati meliputi kadar trigliserida, albumin, dan fosfor darah. Pemeriksaan kadar trigliserida, albumin, dan fosfor secara berturutan menggunakan metode Gliserol Fosfo Oksidase-Fenol 4-Aminoantiprin (GPO-PAP), photometrics bromocresol green, dan cypress diagnostic. Data profil metabolit darah terhadap kinerja reproduksi dikorelasikan menggunakan model Pearson correlation. Hasil penelitian menunjukkan bahwa koefisien korelasi trigliserida, albumin, dan fosfor terhadap PPM dan S/C masing-masing sebesar-0,521 dan -0,650; -0,487 dan -0,612; -0,513 dan -0,700. Dapat disimpulkan, terdapat korelasi yang nyata bernilai negatif antara kadar trigliserida, albumin dan fosfor terhadap PPM, dan korelasi sangat nyata bernilai negatif antara kadar trigliserida, albumin dan fosfor terhadap S/C.

Kata kunci: metabolit darah, sapi peranakan Friesian Holstein laktasi, kinerja reproduksi

INTRODUCTION

Low reproductive performance is one of the reproduction problems among dairy cattle in Indonesia. Blood metabolite profile might be related to cattle fertility status and influences their reproductive performances. Hence, there is a need to investigate the influence of blood metabolite profile on the reproductive performance of Holstein Friesian crossbred (HPC) cows. Reproductive performance of dairy cattle can be examined from various parameters such as post-partum mating (PPM) and service per conception (S/C). Low reproductive performance is indicated by high S/C, low conception rate and long calving interval (Wahyudi *et al.*, 2013). Nutritional deficiency and imbalance have been reported to inhibit reproductive system and to reduce reproductive performance (Guzel and Tanriverdi, 2014). Nutritional deficiency influences anterior hypophysis function, reduces follicle stimulating hormone (FSH) and luteinizing hormone (LH) productions and secretions due to lack of adenosine triphosphate (ATP), resulting in ovarian failure (Prihatno *et al.*, 2013).

Dairy cow has high metabolic requirement that must be met through appropriate feeding. Inadequate feed quality and quantity will reduce glucose and mineral concentration in the blood. Sufficient glucose and albumin concentration can support synthesis and secretion of gonadotropin releasing hormone (GnRH), FSH, LH, and other reproductive hormones to support normal reproductive performance in cattle (Wu, 2010). Low triglyceride level is insufficient to fulfill the energy needs of dairy cattle, resulting in abnormal estrus behavior. Albumin plays an important role as a carrier of several substances in the blood and is also act as a source of amino acids for protein synthesis. Amino acid deficiency results in the disturbance of gonadotropin and gonadal hormone biosynthesis, resulting in weak estrus symptoms, calm estrus, repeated anestrus mating, early embryonic death and stillbirth (Agrawal *et al.*, 2015). Phosphorous plays an important role in ATP transfer; if blood phosphorous level drops significantly, phosphorous deficiency can occur. This can cause reproduction disturbance such as anestrus, low conception rate, long calving interval

(CI), early embryonic death, and stillbirth. Phosphorous and calcium plays an important role in reproductive hormone and enzyme regulations at subcellular level in estrus initiation in animals (Butani *et al*., 2011; Kumar *et al*., 2018).

Blood metabolite profile is used to identify nutritional problem among dairy cattle and management to obtain cattle nutritional status profile in order to investigate short and long term effect from the feeding given and to define the optimal needs of dairy cattle. Short term nutritional status of cattle can be described from fecal evaluation, blood metabolite profile and reproductive performance while long term nutritional status is described from body condition score (BCS). Therefore, this study was conducted to investigate the correlation between blood metabolite profiles (triglyceride, albumin and phosphorous) towards dairy cattle reproductive performances (S/C and PPM) in lactating Holstein Friesian crossbred cows raised in smallholder farmers.

MATERIALS DAN METHODS

This study was conducted in smallholder farmers in Cangkringan, Sleman, Yogyakarta from August to November 2018. Blood samples were collected from 20 Friesian Holstein crossbred cows aged 3-5 years old, weight 350-450 kilograms, had BCS 2-3 (out of 5 scales), were in the second to sixth lactation and not pregnant. Reproductive performance was obtained from data recorded by farmers, including PPM and S/C.

Blood sampling

Blood sample was collected once during estrus phase from caudal vein using venoject needle (Terumo, Japan) connected to collection tube (Vacutainer) containing Ethylenediaminetetraacetic acid (EDTA, Merck, Germany). Identification of estrus cycle phase was conducted before blood sampling by vaginal smear analysis. Other data obtained were body weight, pulse rate, respiratory rate and body temperature to know whether the cow were in a normal physiological condition or not. Blood samples were centrifuged at 3000 rpm for 10 minutes, the plasma was then transferred into1.5 mL microtube (Merck, Germany) using syringe (Terumo, Japan), and then analyzed at the Integrated Research and Testing Laboratory of Universitas Gadjah Mada (LPPT UGM). Triglyceride level was measured using glycerine phosphate oxidase-phenol-4-amino antipyrine (GPO-PAP), albumin level was measured using photometric bromocresol green method while phosphorous level was measured using Cypress Diagnostic method.

Data Analysis

Data was analyzed descriptively to investigate the average level of triglyceride, albumin and phosphorous. Further correlative analysis was conducted to investigate the correlation between triglyceride, albumin and phosphorous towards reproductive performance (PPM and S/C). Sugiyono (2007) stated that data analysis using Pearson correlation results in interval scale or ratio, namely 0.001 to 0.200 for very weak correlation, 0.201 through 0.400 for weak correlation, 0.401 to 0.600 for good correlation, 0.601 to 0.800 for strong correlation, 0.801 to 1.000 for very strong correlation.

RESULT AND DISCUSSION

Average S/C from 20 HFC cow samples in smallholder farmers at Cangkringan, Sleman, Yogyakarta were 2.15±0.81 (Tabel 1). Nuryadi and Wahjuningsih (2011) stated that normal S/C value was 1.6 to 2.0. Pramono *et al*. (2008) reported an average S/C of 1.98. Iswoyo and Widiyaningrum (2008) stated that S/C value above 2 causaed failure to achieve ideal calving interval and thus reduced reproductive performance which is economically detrimental to farmers.

Female cow fertility is influenced by internal factors such as reproductive health and animal management. Another factor that influences S/C value was inseminator's skill. Soeharsono *et al*. (2010) added that farmer's knowledge and skill to detect estrus also influenced S/C value. Detection of estrus, knowledge on optimal time for insemination and timely reporting are very beneficial for the success of artificial insemination.

Average PPM from 20 HFC cow samples in smallholder farms at Cangkringan, Sleman, Yogyakarta was 133.65±18.81 days (Tabel 1). Fanani *et al.* (2013) reported an average PPM of 63.77±25.61 days. The majority of dairy cattle were artificially inseminated after parturition, usually at first estrus which was around 60 days. It was suggested that insemination should be done 60-80 days after parturition because it took 50-60 days to complete uterine involution in cows. Factors that influence duration of PPM in cows are nutritional content of feeds, time to detect estrus by farmers and hormonal factors (Widayati *et al*., 2018).

Correlation between Blood Triglyceride with S/C and PPM

The average triglyceride level in HFC cows was 6.02±1.23 mg/dL (Table 1). Normal cow blood triglyceride level was <150 mg/dL and it was considered to be high when reaches >500 mg/dL (Petkova *et al.*, 2008). Turk *et al.* (2004) stated that normal dairy cattle triglyceride level was 7-30 mg/dL. In agreement to triglyceride level reported by Turk *et al*. (2004), the 20 HFC cows examined had normal triglyceride level.

Analysis results showed that blood triglyceride level had a strong negative correlation towards PPM and S/C (Table 2). Negative correlation in this study showed that if blood triglyceride level increases, the PPM and S/C will decrease. Hence, abnormal triglyceride level will reduce PFH cow reproductive performance by lengthening PPM and increasing S/C number. Strong correlation showed that blood triglyceride strongly impacts PPM and S/C.

Strong correlation coefficient shows the strength of correlation between triglyceride level with PPM and S/C. Triglyceride level is highly influenced by fat and carbohydrate content of feeds (Petkova *et al.*, 2008). Triglyceride is obtained from fats (saturated and unsaturated fatty acid) and carbohydrates (simple and complex) of feed. Triglyceride was used as energy source and the excess would be stored inside liver (Soehardi, 2004 as cited in Faza *et al.*, 2017). Several studies showed that cows fed with sufficient calories feed grew faster and showed normal estrus symptoms (Son *et al*., 2001 as cited in Romano *et al*., 2005).

Correlation between Albumin with S/C and PPM

Analysis result showed that blood albumin level was strongly correlated with PPM and S/C (Table 2). Negative correlation in this study meant that the higher the albumin level, the lower the PPM and S/C value, resulting in higher reproductive performance. Analysis result also indicated that blood albumin level has a significant correlation towards S/C. Reduction of blood albumin levels involves multifactorial processes which include albumin synthesis and destruction, extra vascular leaks, and protein intake (Ballmer, 2001). Ellah *et al.* (2010) stated that there was no difference in albumin levels during estrus cycle and no significance on large follicles. Positive correlation was found between albumin levels inside follicular fluid with follicular size. Albumin could bind chemicals and minerals inside follicular fluid to support follicular growth and maturation.

The main factors that influence albumin synthesis are feeds protein content, colloid osmotic pressure, hormonal actions (thyroid and glucocorticoid hormones) and diseases. Albumin synthesis inside the liver is influenced by amino acids supply from protein contained in the feeds. High albumin level showed the need for amino acid and protein for biosynthesis GnRH and LH important for ovulation initiation (Amle *et al.*, 2014). Low albumin level caused reproductive hormone disturbance, resulting in inactive ovary (Khan *et al.*, 2010).

Correlation between Phosphorous with S/C and PPM

Analysis result showed that blood phosphorous level had a strong negative correlation with PPM and S/C (Table 2). Negative correlation in this study meant that the higher phosphorous level, the lower the PPM and S/C value. Data analysis also revealed that blood phosphorous level significantly influenced S/C. Ali *et al.* (2014) found that Ca : P ratio of 1.5 : 1 would improve reproductive performance such as increase in conception rate (CR) and reduction in CI. There was a significant correlation between fertility and calcium phosphorus ratio in feed. Cows fed with low phosphorous feed experienced ovarian failure and fertility reduction.

Table 1. Average reproductive performance and blood triglyceride, albumin, and phosphorous level in lactating PFH cows

Cow's code	Postpartum mating (days)	Service per conception (times)	Triglyceride (mg/dL)	Albumin (g/dL)	Phosphorous (mg/dL)
1	153	1	6.3	4.14	7.88
2	155	3	5.2	3.60	6.71
3	122	2	7.9	4.00	8.68
4	141	2	8.0	4.00	7.47
5	166	3	5.1	3.83	6.89
6	132	2	5.2	3.74	7.43
7	120	1	7.6	3.83	7.86
8	135	3	4.1	3.75	7.06
9	115	2	7.1	4.36	7.43
10	147	3	5.8	3.43	6.31
11	139	1	6.3	4.26	8.28
12	100	1	7.1	4.4	9.62
13	130	3	5.3	3.68	6.93
14	157	3	3.5	4.14	6.6
15	120	2	5.1	4.3	7.03
16	127	2	7.1	4.15	7.86
17	98	1	6.8	3.93	6.99
18	150	3	5.1	3.25	7.31
19	148	3	5.7	3.54	6.21
20	118	2	6.1	4.35	7.56
Average	133.65±18.81	2.15±0.813	6.02±1.23	3.94±0.33	7.40±0.82

Table 2. Correlation matrix between variables

	S/C		PPM	
Triglycerides	r= -0.650	p= 0.002**	r= -0.521	p= 0.018*
Albumin	r= -0.621	p= 0.004**	r= -0.487	p= 0.030*
Phosphorous	r= -0.700	p= 0.006**	r= -0.513	p= 0.021*

*Different superscripts within the same column indicate significant difference (P<0.05)
**Different superscripts within the same column indicate significant difference (P<0.01)
r= Correlation coefficient, p= Significance value

Low blood phosphorous level caused reproductive disturbance such as anestrous, low conception, long calving interval, embryonic death, stillbirth, and delayed sexual maturity (Chaudhary and Singh, 2004). Studies showed that phosphorous intake of 70-80% of normal requirement caused a significant reduction in fertility rate (S/C 3.7) (Yasothai, 2014). Ali *et al.* (2014) found that blood phosphorous level dropped significantly in cows with anestrous and repeat breeding. This might be explained by the role of phosphorous in ATP transfer. Mineral deficiency and disturbance of main biochemical parameters caused a decreased immunity and productivity, disturbed health status, and reduced reproductive performance (Cerutti *et al*., 2018).

CONCLUSION

There are significant negative correlation between triglyceride, albumin, and phosphorus levels with PPM in the lactating HFC cows. Triglyceride, albumin, and phosphorous levels also had significant negative correlation with S/C.

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