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Factors Associated with the Odds of Pregnancy for Dairy Cattle after Treatment of Ovarian Disorders in Northern Vietnam

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ABSTRACT

The study was carried out to assess factors related to the odds of pregnancy for dairy cattle after treating ovarian disorders in some Northern Vietnamese provinces. A total of 449 ovarian-disordered heifers and cows were given appropriate treatments for their diseases and were diagnosed by rectal palpation with pregnant status after artificial insemination. Data collected from ruminants were analyzed by applying Binary Logistics Procedure in SAS 9.0 and three methods were utilized to test the significant level of regressive coefficients related to the factors, which included various nutritional, environmental, and management factors (locations, parities, ages, body condition scores, housing types, mineral licking block diets, rubber bedding, and categories of disordered ovaries). Based on the odds ratio, cows and heifers in Ha-Noi zone had higher odds of pregnancy than in Vinh-Phuc province (odds ratio [OR] =1.97, p<0.05). Multiparous cows were likely to get a lower chance to conceive than heifers. The odds of pregnancy for dairy cattle decreased with the increase of age, and with each month increased of age, the odds of pregnancy decreased by 1.8%. The better housing condition the dairy cattle live in, the higher pregnancy rate they get. The pregnant opportunity of the dairy cattle kept in the concrete floor house is higher than that kept in rubber floor house (OR=4.03; p<0.0001). Adding mineral licking block in their diet affected the conception odds but did not affect BCS. When the appropriate treatment was used, dairy cattle with inactive ovaries were less likely to conceive than those with cystic ovarian disease and persistent corpus luteum.

Keywords: dairy cattle; ovarian disorder; pregnancy

INTRODUCTION

Ovarian diseases were the important issues affecting reproductive performances in dairy herds (Jeengar *et al.*, 2014; Cattaneo *et al.*, 2014; Long *et al.*, 2017a; Hassan *et al.*, 2017; Bors & Bors, 2020). They prolong the calving interval, reducing pregnancy rate, milk production, and increasing the culling rate. Many studies have been published to evaluate the effectiveness of treatments for ovarian diseases in dairy cattle, including reproductive hormones such as Gonadotropin-Releasing Hormone (GnRH), Prostaglandin F2α (PGF2α), and intravaginal progesterone releasing devices (CIDR, PRID). However, the effectiveness of treatment was not similar between herds as well as among individuals in the same herd (Bryan *et al.*, 2013; Voelz *et al.*, 2016; Besbaci *et al.*, 2020).

In Vietnam, several previous studies have assessed the status of ovarian diseases in dairy cows (Long *et al.*, 2014; 2017a; 2017b; 2017c) and applied hormone therapy (GnRH, PGF2α, Oestrogen, and intravaginal progesterone releasing devices) in treatments, but their effects were different. Long & Thuy (2015) surveyed 156 cows with delayed estrus in Bavi district (Ha-Noi capital, Vietnam) and showed that the rate of ovarian disorder incidence reached 62.82%, the pregnancy rates of cows after treatment of persistent corpus luteum (PCL), cystic ovarian disease (COD), and inactive ovary (IO) were 87.71%, 85.71%, and 88.89%, respectively. The results from Luu *et al.* (2014) showed the rate of ovarian diseases was 16.75% (125/746 cows), the pregnancy rates after treatments of PCL, COD, and IO were 72.72%, 71.42%, and 61.76%, respectively. Some studies reported that the effectiveness of ovarian disease treatment was related to biological and environmental factors (Kim & Jeong, 2019) such as body condition score (BCS), time-point of AI, transition period, heat stress, parity, age, and days open (Siddiqui *et al.*, 2013). In each farming condition, the treatment effect was different (López-Gatius & Hunter, 2019; Lima *et al.*, 2012). Therefore, it is necessary to identify the factors related to the pregnancy rate in dairy herds with ovarian diseases under different farming conditions to improve the success rate of the treatment protocols. Bringing cows back to the normal cyclicity and increasing the chances of pregnancy after treatment would help cows get pregnant sooner again, shorten the calving intervals, and increase milk yield. Thereby, all mentioned above, they found the scientific basis for developing suitably treated protocols for cow's condition and farming environment. Therefore, the study was carried out to assess factors related to the odds of pregnancy for dairy cattle after treating ovarian disorders in some Northern Vietnamese provinces.

MATERIALS AND METHODS

Animals

As many as 449 dairy cows and heifers with over 87.5% Holstein Friesian in some dairy households in Ha-Noi capital (21°1'42.6396''N latitude and 105°48' 17.3412'' E longitude) and Vinh-Phuc province (21°19'59.99"N latitude and 105°34'0.01" E longitude) were observed from January 2017 to December 2018. The dairy cattle were all delayed in estrus, including 107 heifers over 15 months old, and 342 cows had delayed estrus from 90 days or more after calving. Ovaries were examined by rectal palpation for diagnoses. The cows were milked twice a day and housed entirely. The process of conducting research to ensure animal welfare was strictly applied according to the assessment of Vietnam Animal Welfare Association No. 2019-01/QĐ-VAWA.

Rectal Palpation

The method of determining ovarian diseases was conducted according to Long *et al.* (2014) (Table 1) combined with information on the closest estrous day obtained from the farmers. Ovarian disorders were divided into 3 types based on rectal examination of ovaries and classification of diseases by local veterinarians: persistent corpus luteum, cystic ovarian disease, and inactive ovary.

Methods to Evaluate and Categorize Factors Affecting the Pregnancy Rate

Body condition score (BCS) was determined visually at the ovarian examination by local veterinarians, using a scale system from 1 to 5 (1: too thin; to 5: too fat), two consecutive scales separated by 0.25. Normal cows have BCS from 2.75 to 3.0 and fat cows have BCS from 3.25 or more (Ha *et al.,* 2018).

There are two housing types: (1) Well-invested house: built according to the standard model (Good ventilation and hygienic condition); (2) Simple house: a salvaged house, simple design, lowly stable conditions, the roof was a cottage, fibro cement or tiled with low height, poor ventilation. While for bedding types, the floors were divided into 2 types: (1) No rubber and (2) With rubber. For mineral licking block (MLB), there were two types: (1) MLB: Daily the ruminants were licked freely with MLB, based on their demands; and (2) non-MLB: No available MLB in the house.

Table 1. Method to identify ovarian disorders in heifers and dairy cows

Ovarian examination results	Day 1	Day 7-10	Diagnostic conclusion
Corpus luteum	+	+	Persistent corpus luteum
	-	+	Normal
	+	-	Normal
Follicles	+	+	Cystic ovarian disease
	-	+	Normal
	+	-	Normal
Follicles and corpus luteum	-	-	Inactive ovaries

Notes: + = Positive; - = Negative.

Treatment Protocols for Ovarian Diseases

Cows with ovarian diseases were treated by the same veterinarian who examined the ovaries. Cows diagnosed with PCL were treated by protocol 1; Cows with IO and COD were treated by protocol 2. As can be seen in Figure 1, on the first day, cows were injected with 1ml of PGF2α to lysis the functional corpus luteum (Ovuprost, Bayer, Vietnam; IM), and estrus was monitored for the next 7 days. If cows do not express signs of estrus, the injection of 1ml of PGF2α (Ovuprost, Bayer, Vietnam; IM) was repeated again on day 14. AI was performed when the cows were in heat. Figure 2 showed that on the first day, cows were injected with 5 mL of vitamin ADE (Vigantol® - E, Bayer, Vietnam; IM), and 1ml of GnRH (GnRH-1; Ovurelin, Bayer, Vietnam; IM) combined with 1.38g progesterone (Controlled Internal Drug Release - CIDR - EAZI-breed™ CIDR®, Pfizer Animal Health, New Zealand). On day 7, the CDR was withdrawn and the cow was injected with 1ml of PGF2α (Ovuprost, Bayer, Vietnam; IM) and the second injection of GnRH (GnRH-2; Ovurelin, Bayer, Vietnam; IM) was conducted after the next 24 hours. Estrus was detected for the next 5 days, and when the cows were in heat, AI was performed by local veterinarians who were highly skilled and had more than 10 years of experience. Pregnancy was diagnosed by rectal palpation from day 60th after AI.

Data Analysis

Using the bivariate Logistic regression analysis method in SAS 9.0 to analyze data, 8 different regression models have one general factor as follows:

$$Logit(\pi_i) = \beta_{0_i} + \beta_i F_i + e \qquad Logit(\pi_i) = \ln\left(\frac{P=Y_i}{P=J_i}\right)$$

where Logit (π_i) was the i^{th} logistic regression function; β_{0i} was the i^{th} intercept constant of each equation; β_i was the slope of each model of the i^{th} factor; P=Yi was the probability of the event of the i-th independent event (the probability of pregnancy of the cow when inseminated in the treated cycle); P=Ji was the probability of non-event of independent event i (the probability of no pregnant cow when inseminated in the treatment); and e was randomly residual error.

F_i is the i^{th} factor:

1. Model 1: Fixed effects of location: i=2 (Ha-Noi, Vinh-Phuc Province).
2. Model 2: Fixed effect of parity: i=3 (Parity=0 (heifer), 1st parity, 2nd parity, or more).
3. Model 3: Continuous effect of cow age (in the month)
4. Model 4: Fixed effect of housing type: i=2 (Simple house and well-invested house).
5. Model 5: Fixed effect of body condition score: i=3 (BCS1, BCS2, and BCS3).
6. Model 6: Fixed effect of different bedding types: i=2 (with rubber and without rubber bedding).
7. Model 7: Fixed effect of MLB addition: i=2 (Supplemented and non-supplemented cows).
8. Model 8: Fixed effect of the ovarian disorder type: i=3 (PCL, COD; IO).

Parameters of factors such as location, parity, BCS, housing type, bedding type, the addition of MLB, and type of ovarian diseases were treated as the fixed effect; age in months was treated as a continuous variable.

RESULTS

The results in Table 2 showed that different factors were related to the odds of pregnancy of cows after treatment of ovarian disease, including location, parity, age, the addition of mineral licking block, housing type, bedding type, and categories of the ovarian disorder according to Likelihood, Score, or Wald (p<0.05). Exception for BCS, the regression coefficient estimated by all 3 testing methods was not statistically significant (p>0.05).

After treatment for ovarian disorders according to the respective research protocol, the odds of pregnancy after insemination of cows in Ha-Noi was higher than those in Vinh-Phuc (Estimate was positive (0.68); p= 0.0061) (Table 3). The chance of pregnancy after insemination of cows with ovarian disorders after treatment was 1.97 times higher for cows in Ha-Noi compared to those in Vinh-Phuc (p<0.05).

The odds of pregnancy after treatment of ovarian disorders in heifers were higher than that in primiparous and multiparous cows (p<0.05). The chance of pregnancy in heifers after treatment was 3.54 times higher than that in primiparous cows and 2.58 times higher than that in multiparous cows. However, the comparison of the chance of pregnancy between primiparous and multiparous cows was not statistically significant (p>0.05).

As can be seen in Table 3, cows raised in well-invested houses had a much higher odds of getting pregnant after treatment of ovarian disorders than cows raised in the lowly-invested houses or simple houses (p<0.05). It is assumed that a cow transferred from a lowly-invested house to a well-invested house, after ovarian-disordered treatment, the odds of pregnancy increased by 3.44 times higher compared to the cow raised in a lowly-invested house (p<0.05).

Meanwhile, cows raised in a house with a concrete floor had a higher chance of getting pregnant after disordered ovarian treatment than cows raised with the rubber floor (Table 3). According to the results, when cows were transferred from a house with a concrete floor to that with a rubber floor, the chance of getting pregnant was 4.03 times higher than the cows raised in a house with a rubber floor (p<0.05).

Analyzing the effects of diet on ovarian disorders showed that cows supplemented with MLB had a 60.0% chance of getting pregnant than cows not supplemented with MLB. In other words, the chances of getting pregnant were 40% lower for cows that were not supplemented with MLB compared to those with MLB supplementation (p<0.05) (Table 3).

Cows with inactive ovaries had a lower chance of getting pregnant after treatment than cows with cystic ovarian disease (OR= 0.37) and with corpus luteum (OR= 0.59) (Table 3). (p<0.05). There was no statistically significant difference in the chance of pregnancy after treatment between cows with persistent corpus luteum (PCL) and cystic ovary disease (COD) (p>0.05).

The results of Table 4 showed that the chance of pregnancy decreased when the cow's age increased. With the disordered cows after treatment, increasing

Table 2. Testing the effect of factors on the odds of getting pregnancy for heifers and cows after treatment of ovarian disorders (Hypothesis test of coefficient β=0)

Factors	Methods of tests								
	Likelihood Ratio			Score			Wald		
	χ2	DF	Pr> χ2	χ2	DF	Pr> χ2	χ2	DF	Pr> χ2
Location	8.00	1	0.005	7.67	1	0.006	7.51	1	0.006
Parity	18.89	2	<0.0001	17.68	2	0.000	16.69	2	0.000
Age (in month)	7.73	1	0.005	7.84	1	0.005	7.66	1	0.006
Body condition score (BCS)	0.86	2	0.650	0.86	2	0.652	0.85	2	0.653
Housing type	22.36	1	<0.0001	21.11	1	<0.0001	19.85	1	<0.0001
Mineral licking block addition	4.48	1	0.034	4.53	1	0.033	4.49	1	0.034
Bedding type	27.93	1	<0.0001	26.08	1	<0.0001	24.02	1	<0.0001
Categories of ovarian disorders	10.67	2	0.005	10.26	2	0.006	9.98	2	0.007

Notes: χ2= Chi-Square; DF= Degree of freedom.

each month of age will decrease the chance of pregnancy by 1.8% ($p<0.05$).

DISCUSSION

The responses of estrous induction in estrous delayed cows were not similar inter herds or intra herd, and they were seemed to depend on factors such as age, parity, BCS, and result in calving intervals (Rodrigues *et al.*, 2012; Pancarcı *et al.*, 2013; Khalil, 2019). The estrus response after treatment for the ovarian disease was also related to previous anestrus status and days in milk in the herd (Garcia *et al.*, 2013).

Estrus delayed cows in Ha-Noi had a higher chance of getting pregnant than cows in Vinh-Phuc after treatment. This result indicated that the treatment protocols for ovarian disorder were more effective in dairy cows raised in Ha-Noi capital than in Vinh-Phuc province. The cause may be due to the differences in the sub-climate of the locations: the differences in soil, water resources, temperature, humidity, and the intensity of solar radiation. These conditions affected the physiological status of cattle. During the duration of this research (2017-2018), the annual average temperature and humidity in Vinh-Phuc location (about 27-27.5 °C; 79-81.5%) were higher than those in Ha-Noi (about 26-26.5 °C; 72–72.5%). The time with an average temperature above 26 °C was 2 months longer in Vinh-Phuc province than in Ha-Noi capital (summer and autumn). Besides, in Vinh-Phuc, there are about 4 months from March to October with more sunshine hours/month than in Ha-Noi, i.e., from June to September (Vietnam National Center for Hydro-Meteorological Forecasting). With the finding from Wolfenson & Roth (2019), when the air temperature is above 26-27 °C, even a slight increase of 1-2 °C could increase cows' body temperatures, causing heat stress that affects the fertility of cows. Heat stress negatively impacts the activity of hypothalamic-hypophyseal-ovarian axis and follicle quality, affects fertilization and early embryonic development, and increases the risk of embryonic loss in cattle. When using the Ovsynch protocols for estrus induction in dairy cows, the regression of corpus luteum within 96h after PGF2α injection was reduced in heat-stressed cows (84.8% vs 93.5%) (Broes & LeBlanc, 2014). The difference in temperature, long time exposure to the average temperature exceeding the adaptive temperature threshold of dairy cows (≥26 °C), sunshine duration, or risk of heat stress

Table 3. The odds of getting pregnancy of heifers and cows in two different locations

Factors	Parameters	Variables	Estimated value	SE	95% CI		$\chi2$	Pr> $\chi2$
					Lower	*Upper*		
Location	Likelihood	Ha-Noi vs Vinh-Phuc	0.68	0.25	0.19	1.17	7.51	0.006
	OR	Ha-Noi vs Vinh-Phuc	1.97	0.49	1.21	3.21	7.51	0.006
Parity	Likelihood	Par0 vs Par1	1.26	0.31	0.66	1.87	16.58	<0.0001
		Par0 vs Par2	0.95	0.30	0.35	1.54	9.78	0.002
		Par1 vs Par2	-0.32	0.23	-0.78	0.14	1.85	0.174
	OR	Par0 vs Par1	3.54	1.10	1.93	6.51	16.58	<0.0001
		Par0 vs Par2	2.58	0.78	1.42	4.67	9.78	0.002
		Par1 vs Par2	0.73	0.17	0.46	1.15	1.85	0.174
Housing type	Likelihood	WIH vs LIH	1.23	0.28	0.69	1.78	19.85	<0.0001
	OR	WIH vs LIH	3.44	0.95	2.00	5.91	19.85	<0.0001
Bedding type	Likelihood	CF vs RB	1.39	0.28	0.84	1.95	24.02	<0.0001
	OR	CF vs RB	4.03	1.15	2.31	7.04	24.02	<0.0001
Mineral licking block	Likelihood	MLB vs Non-MLB	-0.52	0.24	-0.99	-0.04	4.49	0.034
	OR	MLB vs Non-MLB	0.60	0.15	0.37	0.96	4.49	0.034
Category of ovarian disorder	Likelihood	IO vs COD	-1.00	0.39	-1.77	-0.23	6.51	0.011
		PCL vs COD	-0.48	0.41	-1.29	0.34	1.32	0.251
		IO vs PCL	-0.52	0.23	-0.97	-0.08	5.30	0.021
	OR	IO vs COD	0.37	0.14	0.17	0.79	6.51	0.011
		PCL vs COD	0.62	0.26	0.28	1.40	1.32	0.251
		IO vs PCL	0.59	0.13	0.38	0.93	5.30	0.021

Notes: OR= Odds ratio; SE= Standard Error; CI= Confidence interval; $\chi2$= Chi-Square; Par0= Heifer; Par1= Primiparous or first parity cow; Par2= Multiparous cow; WIH= Well-invested house; LIH= Lowly-invested house; CF= Concrete floor; RB= Rubber Bedding; MLB= Mineral licking block; IO= Inactive Ovary; PCL= Persistent corpus luteum; COD= Cystic Ovarian Disease; Likelihood= Estimated likelihood value.

Table 4. The odds of getting pregnancy of heifers and cows at different ages

Parameters	DF	Estimated value (likelihood)		OR			χ^2	*Pr*> χ^2
		Est	SE	Est	Lower 95%	Upper 95%		
Intercept	1	15.649	0.300	4.782			271.386	<0.0001
Age (months)	1	-0.018	0.007	0.982	0.969	0.995	76.594	0.006

Notes: OR= Odds ratio; Est= Estimated value; SE= Error of estimated value.

between the two locations of Ha-Noi and Vinh-Phuc can affect the pregnancy rate of cows after treatment of ovarian disease in dairy cows.

Results also showed that there was a higher chance of getting pregnant in heifers than in primiparous and multiparous cows ($p<0.05$). The reasons found were related to BCS loss after calving in primiparous and multiparous cows. Acute BCS loss is caused by its greater mobilization of body reserves for milk production and more unbalanced endocrine and metabolic profiles, which negatively affect the resumption of ovarian after treatment. Similar results have been reported by Inchaisri *et al.* (2010), possibly due to a higher risk of suffering from postpartum disease. Higher milk production could result in the elongation of dominant follicles affecting reproduction (Chebel *et al.*, 2004; Wolfenson *et al.*, 2004). In addition, better treatment response in primiparous cows has been found because of their normal length of inter-estrus intervals (Aziz & Abdel-Wahab, 2017) and higher rates of corpus luteum lysis in response to prostaglandin F2 alpha injection compared with multiparous cows (Martin *et al.*, 2011). Herlihy *et al.* (2013) showed no difference in conception rate at first service by parities after estrus, but ovulation rate was higher in primiparous cows. Although some previous studies had shown a decrease in conception rates at the first AI in the first parity, possibly because of more energy demand for growth, however when the environmental and nutritional impacts were controlled, reproductive performance was more improved in primiparous cows compared to multiparous cows (Inchaisri *et al.*, 2010). In our research, many cases of cows with the first parity had an incompletely developed genital system that required outside intervention, leading to an increased risk of genital damage and creating favorable conditions for microorganisms to invade and cause inflammation, increase the risk of postpartum diseases such as metritis, retained placenta, and delayed estrus. The number of cows with more than 2 parities with delayed estrus in this research was low because of the tendency to culling cows that had many calves with delayed estrus to replace new females. Therefore, for cows with parity 1 and 2, it is necessary to examine, detect, and treat metritis and retained placenta in time to increase the pregnancy rate after artificial insemination in dairy cows.

Many previous studies have shown the relationship between age and fertility in cows where the pregnancy rate after artificial insemination increases slightly with the increasing age, high in parity 3-4, then decreases and markedly decreases in cows over 7 years old (Howlader *et al.*, 2019). According to McDougall *et al.* (2001), when treated postpartum anestrus cows, the pregnancy rate and the ability of corpus luteum formation gradually increased with parity, lower in the 2-year-old group than that in the 3-4-year-old group or more than the 4-year-old group. However, our research showed that the chance of pregnancy decreased as the cow's age increased ($p<0.05$). The reason might be that the old cows with ovarian diseases in this research were mainly cows that had not appeared in estrus for a long time and had been artificially inseminated many times without pregnancy. After treatment, the cows had the opportunity to expose estrus. However, the chances of getting pregnant after treatment were lower. These results suggested that the treatment protocols for the old cows with the ovarian disease should be adjusted or those who were too old to have ovarian disorders should be culled.

This study did not find the difference in the chance of pregnancy in cows with different BCSs. However, several other studies have shown a relationship between BCS and the treatment results for ovarian disease in cows. Cows with poor BCS might not ovulate after treatment or show estrus without ovulation and return to anestrus.

In addition, cows raised in well-invested houses, properly designed houses had much higher effectiveness in treating ovarian disorders than cows raised in lowly invested and inappropriately designed houses ($p<0.05$). In the hot season with simple houses, low elevation fibro or tiled roofs, and high temperature will make cows raised in the house at high risk of heat stress. Heat stress causes a critical decline in pregnancy success per insemination (Hansen, 2019; Ramanathan & Kasimanickam, 2021). Dairy cows housed under high ambient temperature condition causes a decrease in the length and intensity of estrus by disrupting ovarian function. Heat stress causes an increase in stress hormones (cortisol, substance-P, and prolactin), leading to a follicular formation and poor oocyte viability; decreased luteal function leading to a decrease in progesterone; and increases isoprostane-8 and prostaglandin F metabolites resulting in a suboptimal intrauterine environment (Ramanathan & Kasimanickam, 2021). In the rainy season or in the house washed several times during a day, the environment in the animal houses tended to be in wet condition. A humid and poorly ventilated environment facilitates microbial development and can increase the incidence of bacterial diseases that affect fertility, such as mastitis and metritis (Asaf *et al.*, 2013; Mohammed *et al.*, 2019). The research recommended that it is necessary to design houses that are more suitable with the cooling system (fan, ventilation, and sprinkler) in the Vietnam context fulfill the physiological demands to increase the efficiency of the activities of functional ovaries.

Usually, the purpose of adding a rubber mattress was to create a comfortable floor for cows in performing behaviors such as walking, standing, and lying down. Thereby limiting the problems of slip damage, hoof erosion, and damage, indirect impact on mastitis and reproductive diseases (Alawneh *et al.*, 2011; Kumar *et al.*, 2017). However, this research showed that cows raised on concrete bedding, when treated for ovarian diseases, had a higher chance of getting pregnant compared with those raised on rubber bedding ($p<0.05$). The actual assessment showed that cows were mainly kept on the concrete floor, rubber mattresses were added to the house floor in the standing, walking, and lying locations of cows. But when the floor is often washed a lot of times a day, the underside of the rubber mattress is always wet, along with feces and urine left under the rubber bedding, which can create favorable conditions for microorganisms to grow and cause lameness and mastitis for cows. The research recommended that the addition of a rubber mattress on the floor should be considered to suit the housing conditions.

Cows supplemented with MLB had a lower chance of getting pregnant than cows without MLB supplementation ($p<0.05$). This result was in contrast to the other studies showing a positive effect of dietary mineral supplementation on fertility in cattle (Yasothai, 2014; Yugal *et al.,* 2013). Minerals have a great influence on ovarian function and the synthesis of reproductive hormones and play a role in preventing problems before calving, including hypocalcemia, mastitis, lameness, and retained placenta that may affect the fertility of cows after calving. However, the effectiveness of mineral supplementation is achieved when the quantity of the supplement meets the mineral requirements of the cow along with age, gestation status, and lactation stage and is in the biologically useful form (R). Yasothai (2014) reported that excessive mineral supplementation would be not good and has the risk of causing negative problems for animal health (Yugal *et al.,* 2013). In this study, farmers often fed MLB for their cows without controlling their intake. There was a significant difference between cows supplemented with MLB and without MLB supplementation. This result showed that it is necessary to carry out more research on the method of mineral supplementation, more research on how to supplement MLB for dairy cows, and ingredients to be suitable for dairy cows in Ha-Noi capital and Vinh-Phuc province.

In this research, the chance of pregnancy after treatment was lower in the cows with the OI ovarian disorders compared to those with the PCL or COD ovarian disorder ($p>0.05$), and there was no difference between the COD group and PCL group ($p<0.05$). Using PGF2α is the most effective method to treat PCL disease, which helps to stimulate the regression of corpus luteum; while using GnRH is an effective method when used in cows with COD (anovulatory follicular cysts) to help the cows to return to a normal cyclic ovarian condition (Jeengar *et al.,* 2014). The combination of GnRH, PGF2α, and PRID or the Ovsynch protocol has also been shown to be effective in treating bovine ovarian disease (Ari *et al.,* 2017; Naglis, 2019). However, the effectiveness of treatment between ovarian disease groups always needs to be adjusted to suit individual cows in different breeding conditions.

CONCLUSION

The odds of pregnancy after treatment of ovarian diseases in dairy cows were associated with factors including location, age, parity, housing, bedding type, mineral licking block supplementation, and ovarian disorder categories, except for body condition score.

CONFLICT OF INTEREST

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