Ovozoa 12 No. 2, August 2023 p-ISSN 2302-6464 e-ISSN 2722-967X
Available at https://e-journal.unair.ac.id/OVZ/index

Original article

Retained placenta in dairy cows living in an all-day cowshed rearing system

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Open access under CC BY – SA license, DOI: 10.20473/ovz.v12i2.2023.71-80
Received March 25 2023, Revised July 6 2023, Accepted August 5 2023
Published online August 2023

ABSTRACT

This study aimed to determine the prevalence and factors that influence the incidence of retained placenta in dairy cows reared under an all-day cowshed housing system at KUD Tani Wilis, Sendang District, Tulungagung. This study used 216 dairy cows, that had already calved, that had been identified to have medical and reproductive records for data exploration. Data collected included the sex and weight of calf, single or twin calves, age of the dam, cowshed structure (floor, puddles on the floor, gutter, distance of cowshed to waste disposal, type of floor and roof, and sunlight exposure), feed of the dam (forage, concentrate, mineral supplements), frequency of retained placenta cases on calving intervals and days open of the dams. The prevalence of retained placenta was presented descriptively. Factors related to the retained placenta and its effect on calving interval and days open were analyzed by Chi-square test using Statistical Product and Service Solution (SPSS) software version 20 for Windows. The result showed that the prevalence rate of retained placenta was 19.91% of all dairy cow populations in the district. Furthermore, the sex of the calf, age of the dam, sunlight exposure, and the quantity of feed (forage and concentrate) were related to the retained placenta ($p < 0.05$). In addition, retained placenta increased along with extending of days open and calving interval. It could be concluded that the factors that influenced the prevalence of retained placenta were the sex of the calf, the age of the cow, the feed and concentrate, and the cowshed that is not facing the sun.

Keywords: age of the dam, calving interval, days open, gender of the calf, sunlight exposure

INTRODUCTION

Dairy farming is a crucial business contributing to the world economy as it has increasingly been run by smallholder farmers and industry. Henchion *et al*., 2022 argued that milk is an economic commodity with strategic value due to the increasing demand year-by-year. This increasing demand can be achieved by maintaining the reproductive health of cows, including reproductive health, in hopes of good productivity (Khan *et al*., 2016). One of the reproductive cases that must be prevented is retained placenta, as it is profitably

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disadvantaged by delaying the upcoming conception (Kamel *et al*., 2022).

Placenta is retained when the physiological dislodgement of the fetal-maternal connection of the placenta has failed to occur, remaining in the cows' uterus for more than 12-24 hours postpartum (Mahnani *et al*., 2021a). Consequently, when this case is not appropriately treated, it will cause a reduction of reproductive efficiency from the increase in services per conception, reduction in fertility rate to longer days open and calving intervals. The highest economic losses caused by retained placenta were due to low fertility and decreasing milk production (Mahnani *et al*., 2021b). Moreover, the retained placenta can cause delayed uterine involution, ovarian cysts, chronic endometritis, and pyometra, lengthening the calving interval, days open and services per conception (Kashima and Ngou, 2021). Meanwhile, the physiological process of milk production is highly dependent on the continuity of the reproductive cycle. Reproductive disorders as a continuation of placental retention will be followed by a decrease in reproductive performance and milk production (Gohary and LeBlanc, 2018). One of the efforts to prevent high cases of retained placenta is to investigate the key causative factors (Gonçalves *et al*., 2019).

In large-scale pasture-raised dairy farms, retained placenta can be diagnosed seven days before parturition by detecting purines and autophagy (Li *et al*., 2022). The aetiology of retained placenta is influenced by maintenance efforts on hygiene and management factors (Islam *et al*., 2013), physiological and pathological status of dairy cows, namely age and parity of cows, nutrition, and condition of calves (stillbirth, single calving and twin calving) (Mahnani *et al*., 2021b). Studies on the factors that influence the incidence of retained placenta in dairy cows in smallholder farmers have not been found much. This study aims to determine the main factors that influence the incidence of retained placenta in dairy cows kept in cowsheds by smallholder farmers in Sendang, Tulungagung, Indonesia.

MATERIALS AND METHODS

The study was conducted at a rural cooperative named KUD Tani Wilis in Sendang District, Tulungagung, which is located at E 111°43' - 112°07' and L 7°51' - 8°18'. The working area of KUD Tani Wilis covers 108.29 km2, comprising 11 villages. Geographically, this area is on the slopes of Mount Wilis, with an altitude of 576-2552 m. Thereby, the farmers keep dairy cows in cowsheds all the time. The average temperature is 22°C, humidity is 85%, and annual rainfall ranges from 1400-1800 mm, with 90-120 rainy days per year (CSA, 2022). Sendang district has the largest number of dairy cows in Tulungagung, accounting for 13,118 heads which supply 52.18% of the milk production of dairy cows in Tulungagung (Sharfina and Yusuf, 2022).

Dairy cows

This study involved dairy cows in the KUD Tani Wilis area as the study population, which was obtained based on purposive sampling. The criteria for the cows as a sample were cows that had already calved and had health and reproductive records. The number of samples was calculated by the formula n = 4P(1-P)/L2, where n is the number of samples of cattle, P is the percentage of cases of retained placenta in the population, L^2 is the error rate of 0.5% (Martin *et al*., 1987). Based on the percentage of retained placenta cases in the population of 16.1% (KUD Tani Wilis data, 2021), a sample of 216 dairy cows were randomly taken. Data were obtained by interviewing farmers and direct observation to cowsheds.

The data collected includes sex and weight of calf, single or twin calving, age of dam, cowshed structure (floor, puddles on the floor, gutter, distance of cowshed to waste disposal, type of floor and roof, and sunlight exposure), feed of the dam (forage, concentrate, supplement minerals), and frequency of retained placenta cases on calving intervals and days open of the dams.

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Data analysis

The prevalence of retained placenta at the KUD Tani Wilis working area was presented descriptively. Factors related to the incidence of retained placenta and their effects on days open and calving interval were analyzed by Chi-square test using SPSS software version 20 for Windows.

RESULTS

This study showed that the prevalence of retained placenta accounted for 19.91% of the population (43 out of 216). The frequency distribution of retained placenta was presented based on the characteristics of the cows (Table 1), cowshed structure (Table 2), feed intake of the cows during pregnancy (Table 3), calving interval and days open (Table 4).

Table 1 Frequency of retained placenta case based on the characteristics of the calf and the dam

	retained placenta (n, %)		significance
	yes	no	
sex of calf			
male	27 (12.50%)	60 (27.78%)	p <0.05
female	16 (7.41%)	113 (52.31%)	
weight of calf			
less than 30 kg	0 (0.00%)	0 (0.00%)	p >0.05
more than 30 kg	43 (19.91%)	173 (80.09%)	
calving			
single	43 (19.91%)	173 (80.09%)	p >0.05
twin	0 (0.00%)	0 (0.00%)	
age of dam			
>7 years/parity >5	27 (12.50%)	9 (4.17%)	p <0.05
<7 years/parity <5	16 (7.41%)	164 (75.93%)	

The rate of significance level with p <0.05 indicates a significant difference.

Table 2 Frequency of retained placenta case based on the cowshed structure

	retained placenta (n, %)		significance
	yes	no	
floor			
sloped 5-10°	13 (6.02%)	64 (29.63%)	p >0.05
flat	30 (13.89%)	109 (50.46%)	
puddles on the floor			
yes	13 (6.02%)	64 (29.63%)	p >0.05
no	30 (13.89%)	109 (50.46%)	
gutter			
yes	12 (5.56%)	16 (7.41%)	p >0.05
no	31 (14.35%)	111 (51.39%)	
cowshed to waste disposal			
≥10 m	34 (15.74%)	135 (62.50%)	p >0.05
< 10 m	9 (4.17%)	38 (17.59%)	
type of floor			
coarse material	28 (12.96%)	98 (45.37%)	p >0.05
cement	15 (6.94%)	75 (34.72%)	

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type of roof			
rooftile	14 (6.48%)	79 (36.57%)	p >0.05
wave asbestos	28 (12.96%)	90 (41.67%)	
tin roof	1 (0.46%)	4 (1.85%)	
sunlight exposure			
yes	13 (6.02%)	18 (8.33%)	p <0.05
no	30 (13.89%)	155 (71.76%)	

The rate of significance level with p<0.05 indicates a significant difference.

The significance level of the sex of the calf and the age of the dam affected on retained placenta was highly remarkable (p <0.05). Dairy cows that gave birth to male calves (12.5%) had a higher frequency of retained placenta than those that gave birth to female calves (7.41%). Older cows (more than seven years old or parity4 more than 5) were more likely to experience retained placenta than younger cows (less than seven years old or parity less than 5), whereas calf weight (less or more than 30 kg) and single or twin calf calvings had no effect (p >0.05) on the occurrence of retained placenta (Table 1).

Cowshed structure, including the slope of the floor, puddles on the floor, gutter, distance between the cowshed and the cowshed waste storage, type of cowshed floor, and cowshed floor had no significant effect on the incidence of retained placenta (p >0.05). In contrast, open areas of cowsheds exposed to sunlight reduced half of the occurrence of retained placenta compared to cowsheds that had no sunlight exposure (6.02% and 13.89%, respectively) with p<0.05 (Table 2).

The quantity of feed (forage and concentrate) affected the occurrence of retained placenta (p <0.05). Cows fed more forage (10-15% bw) and more concentrate (1-2% bw) daily showed lower retained placenta than cows fed less of those. Meanwhile, mineral supplementation in pregnant cows did not affect the occurrence of retained placenta (p >0.05) (Table 3).

Table 3 Frequency of retained placenta case based on a feed of the dam (daily)

	retained placenta (n, %)		significance
	yes	no	
forage			
10-15% bw	10 (4.63%)	16 (7.41%)	p <0.05
< 10% bw	33 (15.28%)	157 (72.69%)	
concentrate			
1-2% bw	11 (5.09%)	17 (7.87%)	p <0.05
< 1% bw	32 (14.81%)	156 (72.22%)	
supplement minerals			
yes	42 (19.44%)	168 (77.78%)	p >0.05
no	1 (0.46%)	5 (2.31%)	

The rate of significance level with p <0.05 indicates a significant difference.

The incidence of retained placenta was associated with extended calving intervals and days open (p <0.05). Cows with calving interval of more than 13 months had higher cases of retained placenta than cows with calving interval of 12-13 months. Similarly, cows with days open of more than three months had higher cases of retained placenta than cows with days open of 2-3 months (Table 4).

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Table 4 Frequency of retained placenta case on calving interval dan days open of the dams

	retained placenta (n, %)		significance
	yes	no	
calving interval			
> 13 months	26 (12.04%)	19 (8.80%)	p<0.05
12-13 months	17 (7.87%)	154 (71.30%)	
days open			
> 3 months	26 (12.04%)	19 (8.80%)	p<0.05
2-3 months	17 (7.87%)	154 (71.30%)	

The rate of significance level with p <0.05 indicates a significant difference.

DISCUSSION

From the calculation, it was found that the prevalence of retained placenta in KUD Tani Wilis was 19.91% of the sample population. This figure is different from other reports from Indonesian and world dairy farms. Santosa (2002) reported that in Sukabumi, Indonesia placental retention reached 4-18% of calvings. Several publications reported that the incidence of retained placenta was 13.4% in Bangladesh (Islam *et al*., 2013), 1.3-39.2% in the United States (Qu *et al*., 2014), and 20.63 ± 0.012% in Yemen (Al-Yamani *et al*., 2021). The differing prevalence of these research papers appeared to be due to multiple factors that might contribute to the incidence of retained placenta. However, none of these reports linked this phenomenon to the all-day cowshed rearing system because they conducted studies in countries with different rearing systems. Although some reports of placental retention in commercial Indonesian dairy farms have been published, these reports did not link the all-day cowshed rearing system with the occurrence of retained placenta. Therefore, this study is the first to show the prevalence of retained placenta in dairy cows under an all-day cowshed rearing system.

Characteristics of dams and calves

This study showed that male calves tend to cause a higher frequency of retained placenta in their dams than female calves. This study showed the same results that the incidence of retained placenta in dams with male calves was higher (23.12%) than in dams with female calves (17.85%) (Al-Yamani *et al*., 2021). However, the effect of calf sex is considered controversial for the incidence of retained placenta (Bernardi *et al*., 2019). This controversy might be due to different maternal factors of the male from female calves that are remains unclear. Instead of calf sex, gestation length makes more sense for contributing to placental retention because gestational age and the postpartum period determine the strength of placental lodgement. Research has shown that a shorter gestation period resulted in lighter offspring and a higher tendency for retained placenta (Rezende *et al*., 2020). It was reported that female calves (27.57 ± 0.54 kg) were lighter than male calves (30.71±0.19 kg) (Dhakal *et al*., 2013). The incidence of retained placenta in dams that calved to a singleton was lower than that of twins (19.67 and 55.55%) (Al-Yamani *et al*., 2021). This is presumably due to the influence of fetal androgenic hormones from the testes of male fetuses, which can affect the incidence of retained placenta (Rezende *et al*., 2020). Androgen hormones produced by the testes negatively affect the hypothalamic-pituitary axis in the release of Follicle stimulating hormone (FSH) and Luetinizing hormone (LH) (Lodhi and Qureshi, 2000). The inhibition of FSH and LH production will have an impact on decreasing estrogen production (Mark and Michael, 2013). Lack of estrogen and high postpartum progesterone were the causes of failure to expel the placenta (Attupuram *et al*., 2016). In this study, older dams tended to be associated with a higher frequency of retained placenta. Parity significantly affected the incidence of retained placenta (Al-Yamani *et al*., 2021). Old dams have a low uterine muscle tone, which can cause

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dystocia (Mekonnen and Moges, 2016). Dystocia is one of the factors causing retained placenta (Raheem *et al*., 2016). Increasing age of dairy cows, more than five years old, was followed by a decrease in the reproductive endocrine system (Skovorodin *et al*., 2019), making it vulnerable to retained placenta (Attupuram *et al*., 2016).

Cowshed structures

The topography of the research area is located on the slopes of mount Willis, causing the land with contours that are not suitable for comfortable and safe grazing of cattle. This condition makes farmers always keep their dairy cows in cowshed all the time and need opportunities to exercise, even walks. Meanwhile, pregnant cows need 1-2 hours of exercise every day to maintain muscle tone and blood circulation for prime cow health (Black and Krawczel, 2019). Lack of exercise is one of the common causes of decreased myometrial contractility, which are factors in the occurrence of retained placenta (Yusuf, 2016).

The unavailability of grazing land also resulted in the unavailability of sunlight for dairy cows in the cowsheds. The only chance for sunlight exposure is an open enclosure facing the sun. In this study, open areas of cowshed exposed to sunlight tended to reduce cases of retained placenta. Sunlight is beneficial for the synthesis of vitamin D in the skin of cows (Weir *et al*., 2017). Serum vitamin D correlated with antioxidant capacity in postpartum cows and is related with cases of retained placenta. Vitamin D also played an important role in calcium absorbtion (Eder *et al*., 2022). Lower serum calcium levels in cows can lead to placental retention due to weakness of myometrial contraction in the third stage of parturition (Strickland *et al*., 2021). In cowshed, where there is not enough open space for sunlight exposure, it is advisable to replace some parts of the roof of the cowshed facing the morning sun with translucent materials.

Feed of the dam

In this study, cows that consumed forage more than 10-15% bw and concentrates more than 1-2% bw showed lower rate of retained placenta. Feed and concentrates are valuable nutrients for basic physiological needs, fetal metabolism during pregnancy, and milk production (Yang *et al*., 2022). During the periparturient period, adequate nutrition is needed related to milk production and the physiological condition recovery period. Nutritional deficiencies can trigger oxidative stress. Oxidative stress throughout periparturient is one of the contributors to retained placenta (Xiao *et al*., 2021). It has been proven that cows that expel placenta normally revealed higher blood selenium and glutathione peroxidase activity, and lower MDA and thyroxine compared to cows with retained placenta (Jovanović *et al*., 2013). Adequate nutrition in quantity and quality is also essential to prevent various reproductive disorders such as retained placenta (Sammad *et al*., 2022). It is necessary to pay attention to fulfilling nutritional needs during pregnancy, cows need more nutrition in sufficient consumption until calving (Noya *et al*., 2019). Lack of feed nutrition could cause retained placenta during the last 6-8 weeks before calving (Yusuf, 2016). This nutritional deficiency can be in the form of mineral and vitamin content in feed. Feed conditions that lack nutrients such as vitamins E, A, and B as well as the ratio of Ca and P can interfere with the body's immunity and increase cases of retained placenta and endometritis (Tucho and Ahmed, 2017).

Calving interval and days open of the dams

In this study, retained placenta tended to be followed by an extention of the dams' calving intervals and days open. Retained placenta that is not handled properly can progress to endometritis, metritis and pyometra. This uterine pathology caused failure of the endometrium to produce prostaglandin F2α (PGF2α), which functioned to regress the corpus luteum (Kumar *et al*., 2014). The persistence of corpus luteum is followed by the very stable concentration of progesterone in the blood. High progesterone

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levels trigger a negative feedback loop to the hypothalamus and anterior pituitary not to release FSH and LH. Without the release of these hormones, there will be no follicular development, no estrogen and no estrus in cows. Retained placenta can delay the emergence of postpartum estrus (Tagesu, 2018). Days open were longer in cows with retained placenta than in normal cows (Temesgen *et al*., 2022). The extended days open will then be followed by an extended calving interval. By suffering from retained placenta, cows will have longer calving intervals than normal cows (Mahnani *et al*., 2021a).

Treatment of retained placenta

Untreated retained placenta can cause permanent infertility in dairy cows due to structural damage and functional disruption of the endometrial epithelium (Li *et al*., 2022). The endometrium produces PGF2α, which is crucial for the continuation of the estrus and reproductive cycles. The endometrium is also important for embryo implantation, placentation, and fetal development until calving (Moraes *et al*., 2018). Structural damage to the endometrium can cause endometrial dysfunction, which results in dairy cows becoming infertile and even sterile (Molina-Coto and Lucy, 2018).

In the study area, generally treatment for retained placenta was carried out by veterinary technicians under the animal health division of KUD Tani Wilis, Sendang District, Tulungagung Regency, by manual removal. The attachment between the cotyledons and the caruncle was manually released with a sterile hand withinin 12-48 hours postpartum. Treatment was continued with the administration of 800 mg sulfamethoxazole and 160 mg trimethoprim or the combination of intrauterine penicillin-streptomycin and multivitamins pro-injection containing vitamins A, D, E and selenium administered intramuscularly. Recently, retained placenta in dairy cows is suggested to be treated with PGF2α in combination with α-chymotrypsin intra-uterine for faster placental detachment, faster uterine involution, decreased risk of postpartum metritis and restore reproductive performance (Amin *et al*., 2023). Good treatment of retained placenta is expected to restore reproductive function and milk production in dairy cows even though days open and calving intervals are extended. However, losses still occur from medical expenses, maintenance costs and lost opportunities to obtain milk from unproductive dairy cows, as well as lost opportunities to get one calf per cow in a year (Mahnani *et al*., 2021a; Li *et al*., 2022). Therefore, it is necessary to carry out corrective actions to evaluate some contributing factors in reducing the prevalence of retained placenta.

CONCLUSION

The prevalence of retained placenta in KUD Tani Wilis was 19.9%, followed by an extension of days open and calving interval. The sex of the calf influenced the incidence of retained placenta in KUD Tani Wilis, the age of the cow, and limited sun exposure as well as the feed and concentrate. Efforts to reduce the prevalence of retained placenta can be done by improving maintenance management.

ACKNOWLEDGEMENT

Thanks to Drh. Didik Isdiyanto for for his guidance in the field, and Pak Herman for his technical assistance.

AUTHOR'S CONTRIBUTIONS

Sayi Datur Rohmah (SDR), Hermin Ratnani (HR), Sunaryo Hadi Warsito (SHW), Rimayanti Rimayanti (RR) , Sri Pantja Madyawati (SPM) , Sri Mulyati (SM), Abdullah Hasib AH)

SDR: compiled ideas, designed the framework, data acquisition, and drafting the manuscript. HR and SHW: supervision data analysis and interpretation, and manuscript draft. RR, SPM, SM, and AH: critically read and revised the manuscript for intellectual content. All authors read and approved the final manuscript.

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CONFLICTS OF INTEREST

The authors dclare that they have no competing interests related this study.

FUNDING INFORMATION

This research was fully funded by the authors

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