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Effects of retained fetal membranes treatments and dry period length on the subsequent lactation in cows - milk yield and somatic cell count

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Abstract. Different treatment strategies in cows with retained fetal membranes (RFM) may affect subsequent lactation in various ways. Also, excessively short or long dry periods (DP) can decrease milk yield (MY), increase the risk of poor udder health, and increase the risk of RFM. This study aimed to evaluate how different treatment strategies of RFM affect subsequent lactation in cows, i.e., MY and udder health determined on the somatic cell count (SCC) bases in milk, expressed as somatic cell score (SCS). A secondary but equally important objective was to analyse the dry period length (DPL) effect on the subsequent lactation in cows. The cows from two herds were divided into three groups: group 1–healthy control; group 2–cows with RFM, removed manually and treated with intrauterine (IU) antibiotics (AB); group 3–cows with RFM, not removed, but treated IU with AB. The DP of cows was divided *post factum* into shortened (up to 46 days), traditional (47 to 70 days), or prolonged (over 70 days). Statistical analyses were performed using linear multiple regression and multivariate analyses. Differences were statistically significant when P <0.05. The effect of different RFM treatment strategies on MY and SCS was evaluated. There were no statistically significant effects of RFM treatments on the MY in the subsequent lactation. However, there was a tendency (P = 0.07) for SCS in standard lactation to be higher in cows in group 2. The highest economic losses, calculated from the decrease in MY, were detected in the same group. Significantly lower MY was observed in cows with a shortened DPL during the first 30 days PP (P <0.05). The DPL did not affect the SCS.

Key words: cows, retained fetal membranes, treatment strategies, dry period length, milk yield, somatic cell count.

INTRODUCTION

Fetal membranes in cows are considered to be retained if they have not been expelled during the first 24 hours after a calf delivery (Guard, 1999; Kimura et al., 2002; Risco & Hernandez, 2003; Maas, 2004; Han & Kim, 2005; Mordak, 2006; LeBlanc, 2007; Sheldon et al., 2008; Könyves et al., 2009; Dubuc et al., 2011; Zobel & Tkalčić, 2013; Gilbert, 2014; Opsomer, 2015). Tucho & Ahmed (2017) indicated that RFM causes significant economic losses, mainly reduced milk yield, worse reproductive performance, and infertility. Therefore, it is important to choose the right treatment strategy to improve animal health and future productivity and avoid animals' early culling. Melendez et al. (2006) pointed out that RFM decreases milk yield, but cow treatment may counteract this decrease. Their study demonstrated that applied monensin capsule before expected parturition decreased the incidence of RFM and improved milk yield in multiparous cows. Goshen & Shpigel (2006) wrote that a 2-week treatment using intrauterine tetracycline for RFM did not affect milk production. However, Bayril et al. (2015) demonstrated that supplementation in feed with Se and vitamin E during the dry period increases Se level in serum, milk yield, and improves udder health (decreasing SCC) in subsequent lactation, but did not decrease the number of RFM cases.

Excessively short or long dry periods (DP) have multiple adverse effects. The DP is considered as the cow's resting period, about 60 days long before calving. Numerous scientific studies have indicated that shortening or omitting the DP may result in improvements in animal health and energy balance, as well as provide economic and management benefits (Grummer & Rastani, 2004; Rastani et al., 2005; van Knegsel et al., 2013; Heeren et al., 2014), and possibly even improve cow fertility in general (Gümen et al., 2005; Pezeshki et al., 2008; Watters et al., 2009). However, this strategy may cause an increase in RFM (Hamidreza et al., 2017). Bachman (2002) and Annen et al. (2004) demonstrated that milk yield is not affected by shortening or omitting the DP, while other studies show that a shorter DP results in lower MY during the first 100 days PP (Rastani et al., 2005; Steeneveld et al., 2014; Hamidreza et al., 2017; O`Hara et al., 2017) and a higher SCC (O`Hara et al., 2019). Van Knegsel et al. (2014) research demonstrated that shortening or omitting the DP reduced the total MY, protein yield, fat and lactose percentage ($P < 0.05$). However, omitting the DP increases the percentage of milk fat and protein, and SCC in milk, compared to a traditional DP (60 days). O`Hara et al. (2020) described that a shortened DP, as well as a prolonged dry period from 80 to 89 days, both reduce milk yield and increase SCC in milk ($P < 0.001$).

In order to accurately assess SCC during a study, these values should be expressed as somatic cell scores (SCS). The Western Canadian Dairy Herd Improvement Services DHI explains that linear scores, or SCS, are used to determine the udder health status of a cow or herd. A very high SCC for at least one cow in a herd strongly affects the herd's average SCC, but the average SCS of the herd is less affected. Thus, the mean lactation SCS is more representative of real milk loss than the average SCC. Dadpasand et al. (2013) described in their study that high SCS is associated with udder health and milk loss and negatively affects dairy cows' fertility and longevity.

The present study hypothesised that different RFM treatment strategies and different dry period length (DPL) could significantly affect MY and SCS in cows during the subsequent lactation. Therefore, the present study aimed to evaluate how different treatment strategies of RFM affect subsequent lactation in cows, i.e., MY and udder health determined on the somatic cell count (SCC) bases in milk expressed as somatic cell score (SCS). A secondary but equally important objective was to analyse the DPL effect on the MY and SCS in the subsequent lactation.

MATERIALS AND METHODS

Animals and Management

The present research was performed in two Holstein's black-and-white breed dairy herds with 650 and 300 cows. Thirty multiparous cows were included in this study: 9 apparently healthy cows with normal expulsion of fetal membranes were randomly selected from both herds, and 21 RFM cows, apparently healthy at the moment when cow grouping was done, 3–8 years old were selected according to the report from a farm veterinarian. RFM cows were examined clinically.

During this study, the average milk yield per cow in a standard lactation was 7,975 kg per year on the first farm and 6500 kg on the second farm, but the average SCC in milk was 197,000 cells mL^{-1} and 248,000 cells mL^{-1}, respectively. On both farms, all management procedures were similar. The cows were fed twice a day, and water was freely available. During the winter, the animals on both farms were head-haltered in a tie-stall system barn, fed silage of maize, alfalfa, clover and grass, prepared in film wrapped rolls, in silage pits and mixed in a feed mixer. Before the silage, the cows were fed concentrates (barley+wheat) and trace elements, rapeseed flour, and salt. During summer, the cows were allowed to graze for twenty-four hours, while during milking, they were fed concentrates with trace elements, salt and rapeseed flour. In their study, Leso et al. (2019) indicated that the keeping system directly affects dairy cows' longevity and udder health status.

The dry period on both farms was planned to be 60 days.

RFM treatment strategies

All study animals (n = 30) were divided into 3 groups depending on the expulsion of fetal membranes and the applied treatment strategy (Table 1). Group 1 was the control. Group 2 was RFM treatment group where fetal membranes were manually easily separable, and group 3 was RFM treatment group where RFM not easily separable. For all treatment group cows, the cotyledon-caruncle junction was gently tested manually for the first two caruncles. Group 2 and group 3 RFM cows were treated with antibacterial preparation-neomycin sulfate 350000 IU and oxytetracycline hydrochloride 500 mg (Gynobiotic bolus; Novartis Animal Health, Slovenia), 3 boluses were inserted into the uterus.

Table 1. Allocation criteria for cows into the study groups during the deliberate grouping

Group of cows	Fetal membranes expelled during 24 h PP	RFM removed manually	Cows treated IU with antibiotics
Group 1 (n = 9)	YES	-	NO
Group 2 (n = 13)	NO	YES	YES
Group 3 (n = 8)	NO	NO	YES

All cows involved in the present investigation were clinically examined in the first 14 days postpartum (PP). RFM cows which increased a rectal temperature ≥ 39.5 °C, showed signs of PP metritis, but still had appetite received systemic antibiotic treatment with ceftiofur hydrochloride (1.1 mg kg^{-1} subcutaneously per day) for 3 consecutive days. In most severe case (cow did not eat, became apathetic), animals received procaine benzylpenicillin (10 mg kg^{-1} intramuscularly per day) for 5 consecutive days. PP metritis was diagnosed when the uterus becomes filled with a large amount of reddish-brown foul-smelling uterine discharge, mostly containing necrotic tissue debris (putrid discharge that corresponds with Sheldon et al. (2006) definitions of uterine diseases).

The reproductive performance of the multiparous cows involved in the study is shown in Table 2.

Table 2. The summary of cows' reproductive performance

The reproduction indices	Group 1 (*n* = 9)	Group 2 (*n* = 13)	Group 3 (*n* = 8)
Days open to the first service, days	79 ± 9.7^{A}	100 ± 14.2	121 ± 28.3^{A}
Services per pregnancy	3.22 ± 0.66	2.25 ± 0.46	1.83 ± 0.47
First service conception, %	$11^{B,C}$	$33^{B,D}$	$50^{D,C}$
Days open, days	187 ± 25.3	132 ± 19.7	163 ± 22.4

^{A,B,C,D} $P < 0.05$; Group 1 – control group; group 2 – cows with RFM, manually removed and IU antibiotics; group 3 – cows with RFM, not removed and IU antibiotics.

Dry period length (DPL)

The DPL of cows (n = 30) was divided *post factum* into the shortened DPL up to 46 days, the traditional DPL from 47 to 70 days, and the prolonged DPL over 70 days. The influence of DPL on MY and SCC was analysed in the control group cows (n = 9), where fetal membranes expelled during 24 h PP and cows with RFM (n = 21). The DPL for each animal involved in the study was obtained from the Agricultural Data Centre Republic of Latvia*. Agricultural Data Centre Republic of Latvia is a State Institution under the supervision of the Ministry of Agriculture that was established in 1997 to aggregate, process and analyse zootechnical, veterinarian and agricultural information in the Latvian Republic with the goal to create a whole state animal and herd register, pedigree information system in compliance with international requirements.

In both herds, dry cow treatment was done using Cloxacillin benzathine 600 mg together with ampicillin trihydrate 300 mg in a long-acting base (Bovaclox DC, Norbrook Laboratories Ltd, Ireland). The cows were clinically healthy before dried off.

Milk yield (MY) and Somatic cell count (SCC)

MY and SCC for the cows in current and previous lactation were obtained from Latvia's Agricultural Data Centre. Both parameters were evaluated for 30 days PP (MY_{30}; SCC_{30}), 100 days PP (MY_{100}; SCC_{100}) and 305 days PP (MY_{305}; SCC_{305}). The difference in MY and SCC were calculated subtracting the previous lactation from the current lactation.

Somatic cell count in milk indicates not only udder health but also the health of the entire cow (Sematovica et al., 2020). In the present study, the optimal threshold for cow SCC was defined as 200,000 cells mL^{-1} based on previous research by numerous scientists (DeGraves & Fetrow, 1993; Harmon, 1994; Hillerton, 1999; Madouasse et al., 2008; Lusis et al., 2010; Petzer et al., 2017; Lusis et al., 2019). All SCC data were transformed into the logarithmic [$\log2 (SCC \times 10^{-5}) + 3$] somatic cell scores (SCS) before statistical comparison. The lactation average SCS was the arithmetic mean of the

*www.ldc.gov.lv

monthly test day SCC from 7 to 305 days after calving. SCC was expressed on a SCS scale from 0 to 9, where (0) ranged from 0 to 24,000 cells mL^{-1}; (1) 25,000–49,000 cells mL^{-1}; (2) 50,000–99,000 cells mL^{-1}; (3) 100,000–199,000 cells mL^{-1}; (4) 200,000–399,000 cells mL^{-1}; (5) 400,000–799,000 cells mL^{-1}; (6) 800,000–1599,000 cells mL^{-1}; (7) 1600,000–3199,000 cells mL^{-1}; (8) 3200,000–6399,000 cells mL^{-1} and (9) 6400,000–12799,000 cells mL^{-1}.

Statistical analysis

Data were analysed using software Stata IC 12.1 (StataCorp LP, 4905 Lakeway Drive, College Station TX77845, USA, version Stata IC 12.1 for Windows) and displayed for each group as a mean ± SEM. Differences between current and previous lactation MY_{30}, MY_{100}, MY_{305}, SCS_{30}, SCS_{100}, and SCS_{305} were analysed using a Wilcoxon matched-pairs signed-rank test. Linear multiple regression (Multivariable analyses) were performed to assess the RFM treatment's impact on the all above mentioned MY and SCS. Multivariate regression analyses of variance (MANOVA) were performed to assess the impact of DPL on the all above mentioned MY and SCS. All effects were corrected for herd influence. Distribution of DPL between herds was compared by Fisher`s exact test. Differences in results were evaluated as statistically significant when $P < 0.05$.

RESULTS AND DISCUSSION

The analyses of RFM treatment strategies' effects on MY_{30} and SCS_{30} showed no significant differences between the study groups ($P > 0.05$). Comparing the current lactation to the previous of each individual cow, MY_{30} in group 2 cows (RFM, manually removed, AB treated) was decreased on average by 98.74 ± 91.19 kg (corrected for herd effect) as compared to the control group as a reference category. In group 3 (RFM, not removed, AB treated) the decrease was on average by 52.89 ± 99.06 kg. Despite a lack of a significant difference in MY, there were considerable losses noticed from an economic point of view (Table 3). For 1L of milk, the average production cost was 0.24 euro in those herds, and the average purchase price was 0.30 euro, resulting in a total loss of 0.54 euro.

Table 3. Milk yield decrease and economic losses in cows with retained fetal membranes during the first 30 days PP

Group of cows	Milk yield decrease per cow, L	Milk yield decrease per cow group, L	Economic losses per cow, euro	Economic losses per group, euro
Group 1 (*n* = 9)	Reference category	Reference category	Reference category	Reference category
Group 2 (*n* = 13)	98.74	1,283.62	53.32	693.15
Group 3 (*n* = 8)	52.89	423.12	28.56	228.48

Group 1 – control group; group 2 – cows with RFM, manually removed and IU antibiotics; group 3 – cows with RFM, not removed and IU antibiotics.

Comparing the current to the previous lactation of each individual cow, SCS_{30} in cows group 2 and 3 was on average 1.42 ± 1.25 and 1.38 ± 1.36 (corrected for herd effect) higher than in the control group cows, respectively ($P > 0.05$).

Continuing to analyse milk yield parameters and udder health in study cows in the first 100 days PP, there were no significant differences between them ($P > 0.05$). Comparing the current to the previous lactation of each individual cow, MY_{100} in group 2 cows was decreased by 328.13 ± 254.04 kg (corrected for herd effect) compared to the control group as a reference category. In cows of group 3, this tendency was weak. Basically, SCS_{100} was increasing between days 30 and 100 PP in both herds; from 3.03 ± 0.56 to 3.74 ± 0.38 in the herd 1 and from 4.01 ± 0.45 to 4.38 ± 0.37 in the herd 2. Comparing cows' groups, the SCS_{100} in groups 2 and 3 were less than one unit higher than the SCS_{100} of the control group. The economic losses for MY_{100} are shown in Table 4.

Table 4. Milk yield decrease and economic losses in cows with retained fetal membranes during the first 100 days PP

Group of cows	Milk yield decrease per cow, L	Milk yield decrease per cow group, L	Economic losses per cow, euro	Economic losses per group, euro
Group 1 (*n* = 8)	Reference category	Reference category	Reference category	Reference category
Group 2 (*n* = 12)	328.13	3,937.56	177.19	2,126.28
Group 3 (*n* = 7)	21.50	150.50	11.61	81.27

Group 1 – control group; group 2 – cows with RFM, manually removed and IU antibiotics; group 3 – cows with RFM, not removed and IU antibiotics.

Further evaluating the effect of RFM treatment methods on MY during the standard lactation period (305-days milk yield), there were no significant differences between the study groups ($P > 0.05$). In contrast, SCS_{305} differences in the standard lactation period in group 2 by 1.51 ± 0.78 ($P = 0.07$), in group 3 by 1.09 ± 0.98 ($P > 0.05$), respectively, were higher than in the control. The variance of MY_{305} results was too high to compare economic losses.

Tucho & Ahmed (2017) indicated that RFM causes considerable economic losses, mainly due to reduced MY and infertility. Furthermore, because there are many different causes of fetal membrane retention, commonly used RFM treatment strategies do not show any significant effects. In principle, management systems must be designed to decrease the occurrence of RFM. This could be achieved by genetic selection, proper feeding during the dry period, and increased activity such as walking (Tucho & Ahmed, 2017).

The dry period is an important phase of a dairy cow's life cycle. During this phase, the cow and its udder are preparing for the next lactation; hence any abnormalities during the dry period will have a negative effect on the cow's health and milk production after parturition. In the present research, data was collected and analysed to understand the effect of the DPL on MY and SCC in subsequent lactations of cows with and without RFM.

In the control group, cows with fetal membranes expelled during 24 h PP, the average DPL was 66 ± 4.84 days (57–101 days), but in cows with RFM, the average DPL was 60 ± 4.38 days. For analysis, the dry period of cows was divided as follows: shortened DPL averaging 30 ± 5.20 days (4–46 days), 20% of cases; traditional or standard DPL averaging 63 ± 1.58 days (47–70 days), 60% of cases; and prolonged DPL averaging 79 ± 4.46 days (71–108 days), 20% of cases. The authors of various articles have defined the dry period of dairy cows differently. No set standard exists, but there are common trends: <49 days is considered a shortened DP or shorter than the conventional DP; a period of 49–70 days is considered a traditional, standard, or conventional DP (Gulay et al., 2003; Kuhn et al., 2006; Pezeshki et al., 2008; Steeneveld et al., 2013; van Knegsel et al., 2014; Sawa et al., 2015; Hamidreza et al. 2017; O'Hara et al., 2017; O'Hara et al., 2019;), and over 70 days is a prolonged DP (Sawa et al., 2015; Hamidreza et al. 2017; O'Hara et al., 2020).

In the present research, the DPL was different between herds (P <0.05) (Table 5). The first herd had more cows with a prolonged DPL (6 out of 12). In contrast, the second herd had more cows with a shortened DPL (5 out of 18). Therefore, the effects of DPL on MY and SCS were statistically corrected for the herd influence.

Table 5. Evaluating dry period length by a herd

Herd	Shortened DPL	Traditional DPL	Prolonged DPL	Fisher`s exact test	Total
1	n = 1	n = 5	n = 6	P = 0.018	n = 12
2	n = 5	n = 12	n = 1		n = 18
Total	n = 6	n = 17	n = 7		n = 30

In the present research, the effect of shortened or prolonged DPL on MY_{30} and SCS_{30} was compared to the traditional DPL. When comparing shortened DPL with the traditional DPL, the cows with shortened DPL had significantly lower MY by 222.45 ± 96.05 kg (P <0.05). In contrast, no differences were found between prolonged DPL and traditional DPL (P > 0.05). O'Hara et al. (2019) mentioned that a 30–day DP might not be long enough for the mammary tissues to adapt to the coming lactation, especially if the DPL is only a few days. Also, in their study, Sawa et al. (2015) indicated that excessively shortened DP in older cows resulted in a daily MY decrease of more than 10% and have a significant effect on udder health during the first 30 days of subsequent lactation. This was confirmed for MY in the present study, but there were not found significant differences in SCS_{30} between the shortened and traditional DPL (P > 0.05).

Evaluating the effect of shortened or prolonged DPL on MY_{100} and SCS_{100} was compared to the traditional DPL. Cows with the shortened DPL continued to show the same tendency as in the first 30 days PP. Respectively, MY_{100} was 385.69 ± 272.26 kg lower than in cows with traditional DPL (P > 0.05). In cows with prolonged DPL, no significant decrease was observed if compared to a traditional DPL. In their studies, O'Hara et al. (2017) pointed out that milk yield during the first 100 days PP was reduced for the shorter 30-day DP cows compared with 60-day DP cows, but there was no significant difference in total MY. This tendency was also confirmed in the present study that cows with the shortened DP period had a lower MY than those of the traditional DP. Even van Knegsel et al. (2014) emphasised in their research that MY in the first 100 days PP was less in cows with a short or no dry period (P <0.01) compared to cows with a conventional DP. This was also confirmed by Steeneveld et al. (2013) study. However, all the studies of the previously mentioned authors were performed in clinically healthy cows. In contrast, this study evaluated milk production and milk SCC in cows with RFM. When evaluating present study SCS_{100}, no significant differences were observed between cows with different DPL ($P > 0.05$).

Further evaluating the effect of DPL on MY_{305} and SCC_{305} in the standard lactation, it was concluded that the shortened DPL negative impact on MY_{305} continued. Respectively, MY_{305} in shortened DP was 914.05 ± 642.56 kg lower than in cows with traditional DPL ($P > 0.05$). However, no significant decrease was observed in cows with prolonged DPL from cows with traditional DPL ($P > 0.05$). This is in line with O'Hara et al. (2020) investigations indicating that MY in the 305-days lactation was low if the DP was 30–39 days long, which corresponds to the present research with shortened DPL. However, our research differed concerning the effect of prolonged DPL. Unlike no difference in the present study, O'Hara et al. (2020) found the milk yield in the 80–89 day group was the lowest ($P < 0.001$). Also, Hamidreza et al. (2017), in their study, described that reducing DPL to less than 45 days had an adverse effect on MY, milk components, and health status. Whereas, Pezeshki et al. (2008) declared that a 28–days DP had no adverse effect on 305–days MY and on the status of multiparous dairy cow health compared with a standard DP (49–days).

When evaluating present study SCS_{305}, neither shortened DPL nor prolonged DPL showed a significant effect ($P > 0.05$). Van Knegsel et al. (2014) and Steeneveld et al. (2013) indicated that there are no differences in SCC between short and conventional DPL in subsequent lactation, which is also confirmed in the present study. However, Kuhn et al. (2006) indicated that prolonged DPL decreases SCC in the subsequent lactation, which has not been detected in the present study.

CONCLUSION

In the present study, the hypothesis on the effects of the RFM treatment strategies and DPL influence on MY and SCS in cows subsequent lactation was partially confirmed.

No statistically significant differences were observed between the various RFM treatment strategies regarding MY_{30}, MY_{100}, and during the standard lactation. Similarly, no significant differences were observed between the various RFM treatment strategies regarding SCS_{30} and SCS_{100}. There was a tendency ($P = 0.07$) for SCS_{305} to be higher in cows with RFM, where it was removed manually, and the IU antibiotics were applied (group 2). The highest economic losses, calculated from the decrease in MY, were also detected in group 2.

Statistically significantly lower MY was observed in cows with a shortened DPL ($P < 0.05$) during the first 30 days PP. The DPL did not affect the SCS.

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