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Customized voluntary waiting period before first insemination in primiparous dairy cows: Effect on milk production, fertility, and health

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ABSTRACT

A customized voluntary waiting period (VWP) before first insemination was tested in 18 commercial dairy herds in Sweden to assess milk production, fertility, and health in primiparous cows expected to be suited for extended VWP. Cow selection for extended VWP was based on 3 criteria in early lactation: (1) the 10% of cows with highest genomic persistency index, (2) cows with a difficult calving or disease during the first month of lactation, and (3) cows with higher yield during d 4 to 33 after calving than the herd average for primiparous cows. Cows meeting at least one of these criteria were randomly assigned to either treatment with an extended VWP of at least 175 d (ExtExt; n = 174, calving interval [CInt] = 16.3 mo) or treatment with a conventional VWP of a maximum of 100 d; (ExtConv; n = 173, CInt = 12.4 mo). Cows not meeting any of the criteria were assigned to the conventional VWP treatment (ConvConv; n = 183, CInt = 12.0 mo). We found no differences in milk yield per day in the CInt between treatments, although 305-d and whole-lactation (WL) milk yields were higher for ExtExt cows (10,371 and 13,803 kg) than ExtConv cows (9,812 and 10,257 kg). Milk yield at the last test milking before dry-off was lower in ExtExt compared with ExtConv cows (24.9 vs. 28.3 kg), but the results showed no difference in dry period length between the treatments. Regarding reproductive performance, the ExtExt cows had a higher first service conception rate (FSCR; 60% vs. 45%) and lower number of inseminations per conception (NINS; 1.67 vs. 2.19), compared with the ExtConv cows. As expected, ConvConv cows had the lowest milk yield (305-d, WL, and per day) in the CInt; however, FSCR and NINS did not differ between ConvConv cows and cows in the other 2 VWP treatments. Disease incidence was higher for cows in the ExtConv compared with the ConvConv treatment, but there was no difference between ExtExt and the 2 other VWP treatments. Further, no difference in the proportion of cows with good udder health or culling rate was detected between any of the treatments, though due to low prevalence, the study lacked power to draw major conclusions on these results. Thus, prolonging VWP for suitable primiparous cows can produce benefits such as improved fertility in the form of higher FSCR and lower NINS, as well as lower dry-off yield, without compromising milk yield or prolonging dry period length.
Key words: extended calving interval, extended lactation, customized lactation, individual adaption

INTRODUCTION

A conventional 12-mo calving interval (**CInt**) has long been considered most beneficial from an economic point of view (Strandberg and Oltenacu, 1989). However, later studies have shown that extended CInt can bring benefits such as improved fertility (Niozas et al., 2019b; Edvardsson Rasmussen et al., 2023a) and lower milk yield at dry-off without compromising milk yield per day in the CInt (Burgers et al., 2021b; Edvardsson Rasmussen et al., 2023b) or the net partial cash flow per cow and year (Burgers et al., 2022). Additionally, extended VWP may lead to less frequent transition periods, as highlighted in recent reviews (Sehested et al., 2019; van Knegsel et al., 2022). These transition periods involve stressful events such as dry-off, regrouping, diet change, calving, and the start of a new lactation; are associated with negative energy balance and decreased immunity (Pascottini et al., 2022); exhibit the highest incidence of disease during the lactation (Bradley and Green, 2004; Ingvartsen, 2006; Växa Sverige, 2023a). Hence, reducing the number and frequency of transitions might be beneficial for herd health. Retrospective studies indicate that longer

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CInt may improve cow longevity (Owusu-Sekyere et al., 2023) and extend productive life (Remmik et al., 2020). However, a problem is that not all cows can maintain milk production for an extended lactation, leading to longer dry periods (Niozas et al., 2019a; Edvardsson Rasmussen et al., 2023b), which in turn may have negative impacts on fertility and health (Roche et al., 2009; Andrée O'Hara, 2019). Due to high individual variation in persistency and in milk yield per day in the CInt, it has been suggested that customizing the voluntary waiting period (**VWP**) before the first insemination for individual cows can decrease the number of transitions on herd level and limit the negative effects in individual cows with less persistent lactations by subjecting them to shorter VWP (Sehested et al., 2019; van Knegsel et al., 2022).

Prolonging the VWP for selected cows has been studied previously, but to our knowledge, not in a random-controlled setting. Burgers et al. (2021a) studied fertility and milk production in 13 commercial herds already practicing customized VWP and found lower conception rate in cows with longer calving to first service interval (**CFI**); however, retrospective studies of extended CFI may have variable reasons for the extension unrelated to VWP. Lehmann et al. (2016, 2017) assessed milk yield performance in 4 herds in which the herd managers selected cows for different lengths of VWP based on criteria such as milk yield in previous lactation (for multiparous cows) or early current lactation, and body condition. They found that first- and second-parity cows could maintain ECM yield per day in the CInt for calving intervals of up 17 to 19 mo, but that dry period length was extended by 3 to 5 d. Further, they identified milk yield in early lactation as a promising indicator of cows suitable for extended lactation. Burgers et al. (2021b) also studied possible predictors of persistency and milk yield per day in the CInt in cows with extended VWP and found that breeding values for persistency and yield during the first 6 weeks of lactation could be useful as predictors of fat- and protein-corrected milk yield per day in the CInt. A high genomic persistency index (**PI**), as defined by the Nordic Cattle Genetic Evaluation (2022), might therefore be an interesting tool for identifying cows suitable for extended VWP. Based on a simulation study on the economics of extended VWP, Inchaisri et al. (2011) suggested that from a fertility perspective, cows with calving difficulties or disease events during early lactation could benefit from extended VWP. The reproductive system is often impaired when an animal is sick, stressed, or needing to save energy (Pascottini et al., 2022); an extended VWP would allow cows more time to recover and regain energy balance, and thereby improve fertility. This information may help to identify cows that are able to maintain milk production during an extended calving interval and benefit from an extended VWP. On the other hand, healthy cows without calving difficulties, with low to moderate PI value, and with low yield may be better suited for a conventional VWP, as they may be expected to have better fertility and might not maintain milk production during an extended lactation, thereby risking a prolonged dry period.

The main aim of this study was to (in commercial dairy herds) compare different milk production, reproductive, and health traits during the first lactation of primiparous cows expected to be suited for extended VWP, but randomly assigned to either a conventional or extended VWP treatment. We also aimed to compare these results with those of cows expected to be suited for, and assigned to, a conventional VWP. The specific objectives of this study were to investigate how VWP would affect milk yield per day in the CInt, milk yield at dry-off and 305-d, whole-lactation (**WL**) yield, dry period length, milk yield before dry-off, number of inseminations per conception (**NINS**), first service conception rate (**FSCR**), insemination period length (**IPL**), estrus intensity, disease incidence, SCC, and culling rate in cows expected to be suited for extended VWP. The hypothesis was that the cows suited for an extended VWP and receiving an extended compared with a conventional VWP would have improved milk production, fertility, and health.

MATERIALS AND METHODS

Study Design, Herd Inclusion, and Description

The study was performed between October 1, 2020, and October 22, 2022, in 18 commercial dairy herds in southern Sweden. Participation in the study was limited to dairy herds matching the inclusion criteria of having more than 9,000 kg average yearly milk yield, having herd size of at least 100 cows, having average CInt of no more than 14 mo (2017), using a system for daily milk recordings, and being located in southern Sweden and connected to the Swedish National Herd Recording Scheme (**SNDRS**). For this study, 14 of the herds selected had participated in a previous study by our research group (Edvardsson Rasmussen et al., 2023a,b), and 4 new herds were recruited. The mean and range of the main herd characteristics for the 18 commercial herds included in the study are presented in Table 1. Timed estrus is not allowed in Sweden, but all herds were enrolled in the Swedish AI program and they were offered and encouraged to conduct genomic testing of all heifers that could potentially be enrolled in the trial. Methods for estrus detection varied across herds: some used activity sensors, others relied on in-line progesterone analyses in milk (Herd Navigator, DeLaval International, Tumba, Sweden), and still others used visual estrus detection or a combination of these methods to detect estrus.

Table 1. Mean value and range (minimum, maximum) of main herd characteristics, herd size, proportion of Red dairy cattle (RDC), Swedish Holstein (HOL), and cross breed or other breeds (cross/other), average yearly yield (kg of milk), calving interval, and reproduction efficiency[1] for the 18 herds included in the study[2]

Herd	Mean	Minimum	Maximum
Herd size (n)	184	99	468
RDC (%)	37	0	91
HOL (%)	54	6	100
Cross/other (%)	9	0	44
Milk yield (kg)	11,450	9,811	13,437
Calving interval (mo)	12.7	11.8	13.8
Pregnancy rate (%)	35	25	45

[1]Reproduction efficiency calculated as calculated as annual herd pregnancy rate (insemination rate × conception rate).

[2]Data from Swedish National Herd Recording Scheme 2019–2020.

Herd recruitment, inclusion criteria, and data collection were similar to those in our previous study, where all cows were randomized to conventional or extended VWP (Edvardsson Rasmussen et al., 2023a,b). Ethical approval (protocol number 5.8.18–10126/2018) was obtained from the Uppsala Ethics Committee for Animal Research (Uppsala, Sweden).

Cow Selection and Intervention

The study enrolled primiparous dairy cows having their first calf between October 1, 2020, and April 1, 2021, with information available from the herd manager and SNDRS regarding calvings, diseases, test milkings, and inseminations (n = 632). Further, cows culled or planned to be culled during the lactation before they were assigned a treatment (n = 21) and 6 cows that the herd managers believed would not maintain milk yield for an extended lactation, and hence were not willing to enroll, were excluded before randomization. Moreover, crossbreds and cows of breeds other than Swedish Holstein or Red dairy cattle (including Swedish Red, Danish Red, and Ayrshire Cattle) were excluded (n = 75). In addition, 17 cows were excluded that were still in their first lactation (had not had their second calf and had not been culled) at the end of data collection on 22 October, 2022. Thus, a final total of 513 cows were included in the study (Table 2).

Cows expected to benefit from an extended VWP were selected based on 3 criteria in the following order (Figure 1 describes this process for the cows with a complete lactation): (1) The first criterion, which was applied to genomically tested cows (78%) in November 2020, was the 10% of genomically tested cows with highest genomic PI as defined by the Nordic Cattle Genetic Evaluation (Nordic Cattle Genetic Evaluation, 2022), which in this case was PI ≥111. The second and third criteria, which were applied to the remaining cows (not genomically tested [22%] or with PI <111 [90% of genomically tested]) before 40 DIM for each cow, were: (2) cows that had a difficult calving (e.g. cows in need of help during calving, twins, or complications) or disease event (any clinical disease reported, whether related to infections, trauma or metabolic causes) during the first month of the lactation, as reported by the herd manager; and (3) healthy cows with a daily milk yield during d 4 to 33 after calving (based on daily yields from the herd milking system on or test milkings) exceeding the herd average for primiparous cows. The average yield for the primiparous cows between 4 and 33 DIM was calculated for each herd before the start and updated once 4 mo into the study. The first cow, based on calving date, for each criterion in each herd was randomized using Slumpgenerator online software (Lejtzén Design, 2020) to either a treatment with extended VWP of ≥185 DIM (**ExtExt**, n = 167) or a conventional VWP of ≤90 DIM (**ExtConv**, n = 169). Once the first random allocation was made, the assign-

Table 2. Number of cows in the ExtExt, ExtConv, and ConvConv treatments following different inclusion and exclusion criteria and combinations of inclusion criteria applied in the data analysis

Item[1]	ExtExt (n = 167)	ExtConv (n = 169)	ConvConv (n = 177)	Total (n = 513)
Breed				
Holstein	121	118	90	329
Red dairy cattle	46	51	87	184
Criteria used for customizing VWP				
Persistency index >111	20	17	—	37
Disease/calving problems	26	31	—	57
Milk yield (d 4–33) >herd average	121	121	—	242
Cows meeting none of the 3 criteria	—	—	177	177
Inclusion criteria				
VWP according to plan	120	139	132	391
No. of inseminated cows	148	159	147	454
Complete lactation	132	134	130	396
VWP according to plan + complete lactation	104	118	116	338
Exclusion criteria				
Still lactating	7	4	6	17

[1]VWP = voluntary waiting period.

ment of subsequent cows followed an alternating pattern. That is, every second cow was assigned to the opposite group of the previous one and so on, to ensure an even distribution of cows in each treatment and herd. Cows not meeting any of the 3 criteria, and hence not expected to benefit from an extended VWP, were all assigned to a conventional VWP of ≤90 d (**ConvConv**, n = 177). One herd lacked cows in the ConvConv treatment because there were no primiparous cows included with a lower than average yield in early lactation. All analyses, except for culling rate and disease incidence, were performed on the per protocol effect, only including cows receiving their planned VWP ±10 d, i.e., CFI of at least 175 d in the ExtExt treatment (n = 120) and CFI of no more than 100 d in the ExtConv (n = 139) and ConvConv (n = 132) treatments (Table 2). Further, in the analysis of the milk production traits, NINS, and SCC, only cows with a complete first lactation that had a second calf before the end of data collection on October 22, 2022, were selected for further analysis. The reasons for this were to get fair comparisons between the treatments regarding the effect of pregnancy, and because many traits could only be calculated for cows with a complete CInt (such as CInt, dry period length, milk yield/CInt day, and NINS). The final number of individuals available for those statistical analyses included 104 cows in the ExtExt treatment, 118 cows in the ExtConv treatment, and 116 cows in the ConvConv treatment (Figure 1 and Table 2). Three cows lacked information about dry-off date and were excluded from the dry period length calculations.

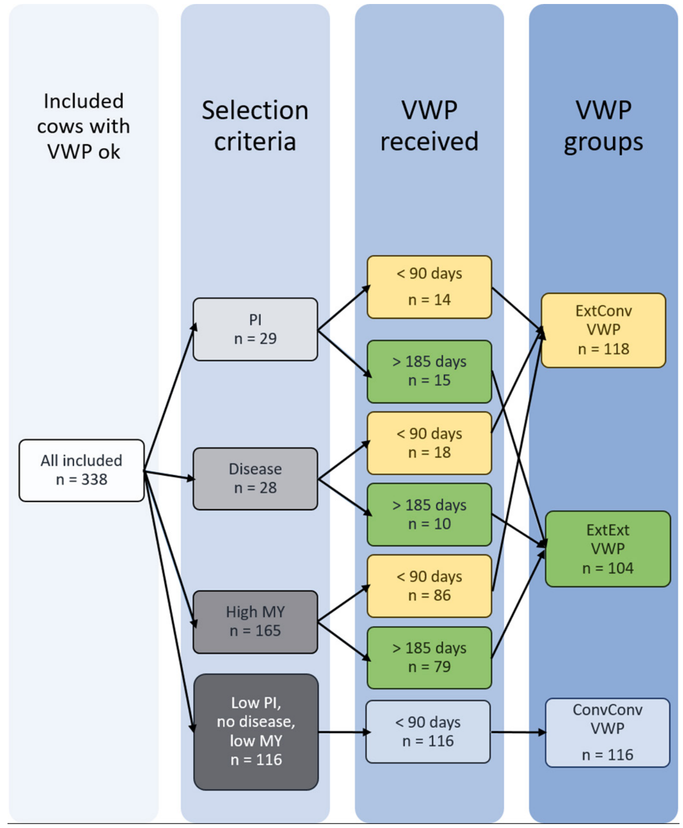

Figure 1. Schematic flow diagram of the study design for cows with a complete first lactation (n = 338), with number of cows allocated to the different voluntary waiting period (VWP) treatments, number of included cows fulfilling at least one of the 3 selection criteria for extended VWP (high persistency index [PI], difficult calving or disease postpartum, and higher than average yield [MY] in early lactation), and number of cows fulfilling each criterion and randomized to and receiving the ExtExt and ExtConv treatments. Cows not fulfilling any criterion for extended VWP were allocated to the ConvConv treatment.

Data Collection, Variable Calculation, and Statistics

Data available from the SNDRS covered the period October 1, 2020, to October 22, 2022, and included parentage, breed, calvings, inseminations, PI, test milking data, SCC, and disease and culling records. Estrus intensity (further described in Appendix Table A1) was available from 12 of the herds (n = 294 cows). Daily milk yield data were collected from the milking systems in the herds. Information about calving difficulties and disease events during the first month after calving was obtained from the herd managers on a weekly to monthly basis until the end of the cow inclusion period. Descriptions and calculations of the variables included can be found in Appendix Table A1.

We used R studio version 2023.3.1.446 (Posit team, 2023) and R software version 4.3.2 (R Core Team, 2022) for data handling and statistical analysis, and GraphPad Prism version 10.0.0 (GraphPad Software, 2023) was used for preparing Figure 2. Mixed models were applied for all analyses, with VWP treatment as a fixed factor (3 levels) and herd as a random factor (18 levels). In the linear and binomial models, breed was included as a fixed factor (2 levels). However, this was not the case in the negative binomial models, as the 2 breeds were not represented in all VWP treatments in each herd. A confidence level of 0.95 was used, results are reported as LSM ± SEM, and *P*-values are reported for the contrasts between the 3 VWP treatments. Linear mixed models were used for all continuous variables and analyzed with the lmer function from the lme4 package in R (Bates et al., 2022). Generalized linear mixed models were used for the binomial variables, fitted by Laplace approximation with the glmer function in the lme4 package in R (Bates et al., 2022). The results from the binomial models are presented as the percentage and proportion of cows (n/N), where n is the number of cows receiving the assigned VWP, conceiving at first service, with SCC <100,000 cells/mL of milk at last test milking, or within each estrus intensity score (Table A1), and N is the total number of cows in each analysis. The numbers of cows with short (<40 d) and long (>70 d) dry period length

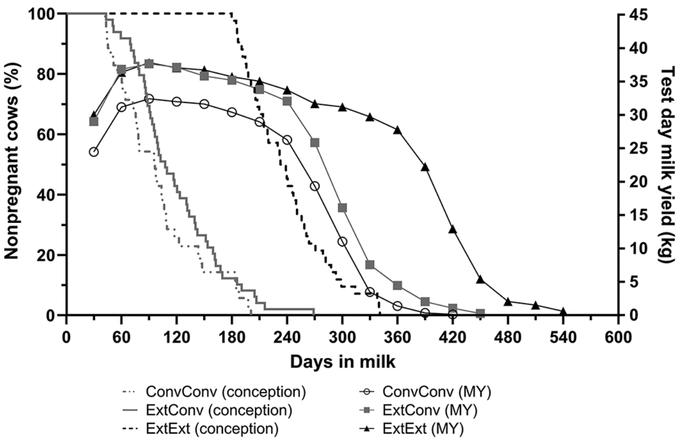

Figure 2. Survival chart showing the proportion of nonpregnant cows per DIM, projected over lactation curves showing kilograms of milk per monthly test milking (TM) after the first calving for cows following the planned VWP treatments (ExtExt, n = 104; ExtConv, n = 118; and ConvConv, n = 116) and with complete lactations. For the lactation curves up to the eighth TM, all cows were still represented in all treatments. After the last recorded TM yield for each cow, subsequent TM milk yields were set as 0 kg to give a correct average yield for each treatment.

(Andrée O'Hara, 2019) were compared with the number of cows with moderate dry period length. Post-hoc tests were done with pairwise comparison, using the Tukey method for *P*-value adjustment with the emmeans function (Lenth et al., 2022). Disease incidence rate from 33 DIM, NINS, and culling rate were analyzed with a negative binomial model, due to presence of underdispersion in the count data. The results were first calculated per herd and VWP treatment, resulting in one value per herd and VWP subgroup (n = 51), with one herd excluded from these analyses because the ConvConv treatment was not represented in that herd. These negative binomial models were analyzed with the glmmTMB function in R (Brooks et al., 2017), using herd as a random factor. The results are presented as the number of disease events and culled cows per 100 cow-years at risk, and for NINS as the number of inseminations of all cows with a complete lactation divided by the number conceptions during the first lactation. Metadata and supplemental files are published in the Swedish National Data Service catalog (Edvardsson Rasmussen et al., 2022), but the full research data cannot be openly published due to restrictions in the agreement with the principal owner of the data (Växa Sverige, Uppsala, Sweden).

RESULTS

Compliance and Milk Production

In the ExtExt treatment, 29% of the cows were not inseminated according to plan, compared with 18% in the ExtConv treatment and 27% in the ConvConv treatment. Of these, 42%, 31%, and 67%, respectively, were not inseminated at all (Table 2). As expected, CInt and lactation length were longer for cows in the ExtExt treatment than for cows in the ExtConv and ConvConv treatments (Table 3 and Figure 2). Moreover, milk yield between 4 and 33 DIM was higher in ExtExt and ExtConv cows (29.5 ± 0.6 and 29.6 ± 0.6 kg) than in ConvConv cows (23.3 ± 0.6 kg; $P < 0.001$). Milk yield from d 4 to 33 did not differ between cows in the ExtExt and ExtConv treatments ($P = 0.95$). Dry period length (measured both as number of days dry and as proportion of cows in different dry period length categories) did not differ between cows in any of the 3 VWP treatments (Tables 3 and 4).

As intended, PI did not differ between the ExtExt and ExtConv groups (103.3 ± 0.9 vs. 101.7 ± 0.8; $P = 0.34$). However, it also did not differ between ExtConv and ConvConv cows (101.7 ± 0.8 vs. 100.6 ± 0.9; $P = 0.59$) or between ExtExt and ConvConv cows ($P = 0.07$).

Whole-lactation and 305-d milk yield (kg milk and kg ECM) were highest for cows in the ExtExt treatment, followed by cows in the ExtConv treatment, and lowest for cows in the ConvConv treatment (Table 3). Daily milk yield and ECM in the CInt were lowest in CovConv cows, but did not differ between ExtExt and ExtConv cows (Table 3). Milk yield at the last test milking (**TM**) before dry-off was 3.4 and 4.9 kg lower in ExtExt and ConvConv cows compared with ExtConv cows (Table 3). Persistency, measured as the change in milk yield per day between the third and eighth TM, was higher in ExtExt cows than in ExtConv and ConvConv cows, but we found no difference between cows in the ExtConv and ConvConv treatments (Table 3). Regarding persistency, measured as change in milk yield per day between the third TM and the last TM before dry-off, we found no difference between the treatments.

Reproductive Performance and Health

By design, CFI was longest in the ExtExt treatment, but it was also longer in ExtConv than in the ConvConv treatment (Table 5). The FSCR was higher in the ExtExt treatment than in ExtConv (60% vs. 45%, $P = 0.04$), but we found no difference between the ExtExt and ConvConv treatments, or between the ExtConv and ConvConv treatments (Table 4). Cows with a complete lactation in the ExtConv treatment needed 2.19 inseminations to conceive, compared with 1.67 for cows in the ExtExt treatment ($P = 0.02$), but we found no differences between ConvConv cows (NINS 1.85) and those in the other treatments (Table 4). A survival chart with the proportion of nonpregnant (open) cows per DIM, illustrating the interval from calving to conception, is presented

Table 3. Total number of cows (n^{tot}) in each analysis, first lactation CInt, lactation length, dry period length, 305-d lactation yield, WL yield, average MY per day in the calving interval, MY at TM 50 to 20 d before DO, and persistency[1] (LSM ± SEM) for cows in the ExtExt, ExtConv, and ConvConv treatments

Variable[2]	n_{tot}	ExtExt (n = 104)		ExtConv (n = 118)		ConvConv (n = 116)		*P*-value		
		Mean	SEM	Mean	SEM	Mean	SEM	ExtConv vs. ExtExt	ConvConv vs. ExtExt	ConvConv vs. ExtConv
CInt (mo)	338	16.3[a]	± 0.1	12.4[b]	± 0.1	12.0[b]	± 0.1	<0.001	<0.001	0.13
Lactation length (d)	335	435[a]	± 4	318[b]	± 4	308[b]	± 4	<0.001	<0.001	0.08
Dry period length (d)	335	61	± 3	58	± 3	59	± 3	0.22	0.58	0.82
305-d MY (kg)	337	10,371[a]	± 210	9,812[b]	± 204	8,146[c]	± 204	0.002	<0.001	<0.001
WL MY (kg)	338	13,803[a]	± 289	10,257[b]	± 279	8,294[c]	± 279	<0.001	<0.001	<0.001
WL ECM (kg ECM)	338	14,458[a]	± 301	10,679[b]	± 291	8,818[c]	± 291	<0.001	<0.001	<0.001
Daily MY (kg/d)	338	27.7[a]	± 0.6	27.3[a]	± 0.6	22.7[b]	± 0.6	0.65	<0.001	<0.001
Daily ECM (kg ECM/d)	338	29.1[a]	± 0.6	28.3[a]	± 0.6	24.1[b]	± 0.6	0.24	<0.001	<0.001
ΔMY TM3 to TM8[3] (kg/d)	338	−0.026[a]	0.005	−0.037[b]	0.005	−0.043[b]	0.005	0.03	0.001	0.39
ΔMY TM3 to TMDO[4] (kg/d)	306	−0.036	0.004	−0.041	0.004	−0.041	0.004	0.32	0.47	0.98
MY TM before DO (kg)	306	24.9[b]	± 1.0	28.3[a]	± 1.0	23.4[b]	± 1.0	<0.001	0.14	<0.001

[1]Persistency measured as average ΔMY in kilograms per day between TM3 and TM8 and between TM3 and DO.

[2]CInt = calving interval; WL = whole-lactation; MY = milk yield; TM = test milking; DO = dry-off; ΔMY = change in MY.

[3]Average change in MY per day between TM3 (the third TM recorded for each cow) and TM8 (the eighth TM recorded for each cow).

[4]Average change in MY per day between TM3 and TMDO (the last TM before DO).

together with the lactation curves for cows in the 3 VWP treatments in Figure 2.

Disease incidence from 33 DIM during the first lactation was higher in the ExtConv treatment compared with the ConvConv treatment (22 vs. 9 cases per 100 cow-years in the study; $P = 0.02$; Table 6 and Appendix Tables A2 and A3). We found no statistically significant difference in culling rate between any of the VWP treatments, either when comparing the per protocol effect (only including cows that received their planned VWP treatment) or the intention to treat effect (including all cows selected and randomized to each VWP treatment), and neither comparison of estrus intensity at first insemination (Table 4) nor SCC at the last TM revealed any statistically significant difference between the VWP treatments. The proportion of cows with an SCC of less than 100,000 cells/mL at their last TM in their first lactation, indicating good udder health (Persson Waller et al., 2020), was 62% (64/104), 58% (69/118), and 65% (75/116) in the ExtExt, ExtConv, and ConvConv treatments, respectively ($P > 0.05$).

DISCUSSION

In this study of customized VWP in 18 commercial herds, primiparous cows expected to be suitable for extended VWP based on high genomic PI, calving difficulties, disease, or higher milk yield in early lactation were randomly assigned to either an extended or a conventional VWP. Cows not meeting any of these criteria were assigned to a conventional VWP.

Milk Production

Milk yield per day in the CInt did not differ between cows in the ExtExt and ExtConv treatments. Moreover, cows in the ExtExt treatment had 6% higher 305-d yield and 35% higher WL yield than cows in the ExtConv treatment. All these results are in line with our previous finding of 6% higher 305-d yield and 28% higher WL yield in cows randomized to extended VWP (Edvardsson Rasmussen et al., 2023b). The higher 305-d yield for the ExtExt cows might be explained by earlier onset of the negative effects of pregnancy on milk production for ExtConv than for ExtExt (Strandberg and Lundberg, 1991; Burgers et al., 2021b). Increased WL yield when the cows had a longer lactation was also expected (Lehmann et al., 2017; Burgers et al., 2021a). The CInt for cows with extended VWP was longer than in our previous study (16.3 compared with 15.2 mo, partially explained by the planned longer VWP of 175 d in the current compared with 145 d in the previous study) which, together with that selection of cows suited for extended VWP to a large part was based on yield, may explain the higher WL yield

Table 4. Total number of cows (n_{tot}) in each analysis, percentage and prevalence (n/N) of cows with short and long dry period, with first service conception, and with a weak, moderate, and strong estrus intensity at first insemination in the ExtExt, ExtConv, and ConvConv treatments

		ExtExt		ExtConv		ConvConv		*P*-value		
Variable	n_{tot}	%	(n/N)	%	(n/N)	%	(n/N)	ExtConv vs. ExtExt	ConvConv vs. ExtExt	ConvConv vs. ExtConv
Short dry period[1]	304	4	(4/93)	4	(4/108)	6	(6/103)	0.998	0.99	0.98
Long dry period[2]	324	11	(11/100)	9	(10/114)	12	(13/110)	0.93	0.73	0.51
FSCR[3]	391	60[a]	(72/120)	45[b]	(62/139)	51[ab]	(67/132)	0.04	0.40	0.48
Estrus intensity[4]										
Weak	294	7	(6/88)	5	(5/100)	2	(2/106)	0.79	0.20	0.43
Moderate	294	45	(40/88)	44	(44/100)	46	(49/106)	0.95	0.73	0.52
Strong	294	48	(42/88)	51	(51/100)	52	(55/106)	0.77	0.99	0.81

[a,b]Proportions within a row with different superscripts differ significantly ($P < 0.05$).

[1]Less than 40 d dry. n = number of cows with short dry period; N = sum of cows with short and moderate dry period length. Moderate dry period length = 40 to 70 d.

[2]More than 70 d dry. n = number of cows with long dry period, N = sum of cows with long and moderate dry period length.

[3]FSCR = first service conception rate; n = number of cows with first service conception; N = total number of cows included in the analysis within each voluntary waiting period treatment.

[4]Weak estrus intensity defined as an estrus intensity score of 0 to 2 reported to the SNDRS; moderate estrus intensity defined as an estrus intensity score of 3 reported to the SNDRS; strong estrus intensity defined as an estrus intensity score of 4 to 5 reported to the SNDRS.

in the present study compared with our previous study (13,803 kg vs. 11,872 kg).

Milk yield per day in the CInt did not differ between cows in the ExtExt and ExtConv treatments, suggesting that customizing extended VWP based on the criteria used in this study can maintain milk yield per day in the CInt for primiparous cows. With a longer lactation and similar dry period length, the proportion of nonproductive dry days was 14% for the ExtExt cows and 18% for the ExtConv cows. Moreover, milk yield at the last TM before dry-off was 12% lower in ExtExt than ExtConv cows, which is also in line with previous findings on the effects of randomized VWP (Niozas et al., 2019a; Burgers et al., 2021b; Edvardsson Rasmussen et al., 2023b). Although milk yield at the last TM before dry-off was 20% higher in ExtConv cows than in ConvConv cows, there was no difference in milk yield at the last TM before dry-off between cows in the ExtExt and ConvConv treatments. This lower yield for ExtExt compared with the ExtConv cows might facilitate dry-off and be beneficial for udder health, as high yield at dry-off has been linked to impaired udder health (Rajala-Schultz et al., 2005). However, even though milk yield at the last TM before dry-off was lower for ExtExt cows than ExtConv cows, the proportion of cows with long dry period (>70d) did not differ between the treatments (11%, 9%, and 12% for ExtExt, ExtConv, and ConvConv, respectively). In contrast, in our previous study with randomized VWP for primiparous cows, we found that 20% of the cows in the extended treatment had a long dry period, compared with 9% of cows with conventional VWP (Edvardsson Rasmussen et al., 2023b). A long dry period (>70 d) may be associated with increased risk of culling, reduced milk yield, and reduced fertility in the following lactation (Andrée O'Hara et al., 2020).

Milk yield per day in the CInt, 305-d yield, and WL yield were all lower in cows in the ConvConv treatment than in ExtExt or ExtConv cows. This was not surprising,

Table 5. Total number of cows (n_{tot}) in each analysis, calving to first insemination interval (CFI), and insemination period length (IPL) of cows with voluntary waiting period (VWP) according to plan and a complete first lactation ending with a second calving, presented as LSM ± SEM of each VWP treatment[1]

		ExtExt (n = 120)		ExtConv (n = 139)		ConvConv (n = 132)		*P*-value		
Variable	n_{tot}	Mean	SEM	Mean	SEM	Mean	SEM	ExtConv vs. ExtExt	ConvConv vs. ExtExt	ConvConv vs. ExtConv
CFI (d)	391	202[a]	± 2	69[b]	± 2	64[c]	± 2	<0.001	<0.001	0.04
NINS	384	1.67[b]	± 0.13	2.19[a]	± 0.14	1.85[ab]	± 0.13	0.02	0.58	0.16
IPL (d)	391	18[b]	± 4	34[a]	± 4	25[ab]	± 4	0.004	0.33	0.18

[a–c]Mean values within a row with different superscripts differ significantly ($P < 0.05$).

[1]Number of inseminations per conception (NINS) was calculated as the number of inseminations per pregnant cow, and the results were calculated per herd and VWP subgroup (n = 51, from 17 herds and 3 VWP treatments, ConvConv, ExtConv, and ExtExt).

Table 6. Total number of cows (n_{tot}) in each analysis, culling rate for all cows randomized to treatments (intention to treat), and culling and disease incidence rate from 33 DIM for cows receiving the planned (per protocol) voluntary waiting period (VWP), per 100 cow-years in the study (time at risk)[1]

		ExtExt		ExtConv		ConvConv		*P*-value		
Variable	n_{tot}	Mean	SEM	Mean	SEM	Mean	SEM	ExtConv vs. ExtExt	ConvConv vs. ExtExt	ConvConv vs. ExtConv
Culling (intention to treat)	500	16	± 4	19	± 4	26	± 6	0.83	0.19	0.48
Culling (per protocol)	384	10	± 3	14	± 4	10	± 3	0.59	0.99	0.59
Disease (intention to treat)	500	11[ab]	± 4	22[a]	± 7	8[b]	± 3	0.09	0.63	0.01
Disease (per protocol)	384	13[ab]	± 4	22[a]	± 7	9[b]	± 3	0.18	0.62	0.02

[a,b]Mean values within a row with different superscripts differ significantly ($P < 0.05$).

[1]The values presented are LSM ± SEM, per VWP treatment (ConvConv, ExtConv, ExtExt).

as only cows with below-herd-average milk yield in early lactation could be assigned to the ConvConv treatment, and milk yield in early lactation between d 4 and 33 was higher in cows in the ExtExt and ExtConv treatments compared with the ConvConv treatment.

Persistency

Regarding persistency measured as reduction in milk yield per day between the third TM and the last TM before dry-off, we found no differences between any of the treatments. However, Burgers et al. (2021b) observed slightly higher persistency for cows with 200-d VWP compared with 50-d VWP. As one of the selection criteria for the cows ExtExt and ExtConv treatments was genomic PI, higher persistency could be expected in both these treatments. However, we found no difference in genomic PI between any of the treatments. A possible explanation is that only the 10% of genomically tested cows with the highest PI were assigned to the ExtExt and ExtConv treatments, and they might have been too few in number to substantially affect the average PI of the treatments. Further, an additional limitation regarding PI was that not all cows were genetically tested, and therefore the 22% of cows not tested might have included cows with high PI. With these limitations in mind, and because it was not an aim of the study to evaluate each separate selection criteria, it is not possible to draw any major conclusions on the effect of this selection criteria from this study.

Fertility

The cows expected to be suited for and receiving an extended VWP (the ExtExt treatment) generally performed better than cows meeting the same criteria but that were randomized to receive conventional VWP (the ExtConv treatment). The ExtExt cows had higher FSCR, and the cows with complete first lactations needed fewer inseminations per pregnancy, and had shorter IPL. These fertility results are in line with findings from randomized studies of extended VWP which report higher FSCR, fewer NINS, and shorter IPL for cows with extended VWP (Niozas et al., 2019b; Edvardsson Rasmussen et al., 2023a). However, the results in the present study are contrary to those in the retrospective study of customized VWP by Burgers et al. (2021b), where cows were grouped by both CInt and CFI. In that study, cows with extended CInt had poorer fertility in terms of FSCR and NINS. When the cows were grouped by CFI, no difference in NINS were observed, although cows with CFI of 140 to 195 d had lower FSCR than cows with CFI of 84 to 139 d. However, the disadvantage of retrospective studies of customized VWP, as also pointed out by Burgers et al. (2021b), is that it is not clear whether longer CFI is due to intentional extension of the VWP or to cows not showing estrus in time, which can affect the results.

The better fertility of ExtExt cows compared with ExtConv cows aligns with results from a retrospective field study, indicating that that high-yielding cows had shorter IPL and lower NINS with increasing CFI (Römer et al., 2020). Further, the better fertility in ExtExt compared with ExtConv cows supports findings by Ma et al. (2022), where cows randomized to extended VWP showed improved cyclicity at the end of the VWP compared with cows with conventional VWP. Further, Nyman et al. (2018) found that cows with an extended VWP also had a greater likelihood of conceiving and maintaining pregnancy. Another possible explanation of the better fertility of the cows in the ExtExt treatment is that diseases in early lactation may have carry-over effects, leading to impaired reproduction over several months, partly due to delayed resumption of cyclicity (Pinedo et al., 2020) or reduced oocyte competence (Ribeiro et al., 2016; Carvalho et al., 2019). Further, as reviewed by Fleming et al. (2018), parental energy status has been found to affect gametes and early embryo development, in both humans and other animals.

We found no difference in estrus intensity at first insemination between cows in any of the treatments in the present study. We cannot explain the discrepancy from the results of our previous study, where estrus intensity was found to be higher in cows with extended VWP (Edvardsson Rasmussen et al., 2023a), but the number of observations in each estrus intensity score group was relatively low in the current study, and therefore the study lacked power to draw conclusions about the lack of difference between the treatments.

Health and Culling

Applying an extended VWP management routine leads to fewer transition periods per unit time. This may lead in turn to a decrease in disease incidence (as discussed by van Knegsel et al., 2022), as disease incidence peaks during the transition period (Ingvartsen, 2006; Växa Sverige, 2023a). As some cows in the ExtExt and ExtConv treatments were selected due to calving problems or disease in early lactation, both those treatment groups might be expected to have higher disease incidence than the ConvConv group. However, this was only seen in the ExtConv group, although disease incidence in the ExtExt group was numerically higher than that in the ConvConv group.

We found no difference in proportion of cows with good udder health, based on SCC of <100,000 cells/mL milk at the last TM before dry-off. We expected to have a slightly higher proportion of cows with high SCC in the ExtExt and ExtConv treatments than in the ConvConv treatment, as those treatments included cows selected due to early lactation disease. However, this was not the case, which might be explained by a weak relationship between health disturbance in early lactation and SCC before dry-off (Kirkeby et al., 2021). Further, the number of sick cows in these treatments was also low and thereby unlikely to influence the proportion of cows with low SCC.

We did not find a difference in culling rate between any of the VWP treatments, either on analyzing the per protocol effect or when including all cows randomized to each VWP treatment, i.e., the intention to treat effect. This was in agreement with findings in our previous study on randomized VWP (Edvardsson Rasmussen et al., 2023a). The culling rate for the cows in the ExtExt and ExtConv treatments in the present study was lower than the average culling rate in Sweden, where the proportion of primiparous cows culled is 26% (Växa Sverige, 2023b). However, as the culling rate was shown to be relatively low, which affects the statistical power and thereby the possibility to draw conclusions on a lack of difference between the treatments.

General Considerations

Because this was a field study in commercial herds, management routines and reporting to the SNDRS varied between herds, as did compliance with the assigned VWP. However, all treatments were represented in each herd (except one herd lacking cows assigned to the ConvConv treatment), and herd was included as a random factor in the statistical models to control for the assumed variation in management and reporting of data between herds.

This study only included primiparous cows; therefore, generalization of the results to multiparous cows, which are generally known to have a less persistent lactation curve (Chen et al., 2024), should be made with caution. However, previous studies including multiparous cows have suggested similar predictors of cows suitable for extended lactations (Inchaisri et al., 2011; Lehmann et al., 2017; Burgers et al., 2021b). Further, also taking into account that persistency differs between individuals, one may speculate that customization may possibly be even more valuable for multiparous cows. Further research validating predictors for this population would be desirable.

The per protocol effect on milk production was explored, only including cows following the planned VWP. In a previous study of randomized VWP, low milk yield was more commonly reported as the reason for culling for cows randomized to extended compared with conventional VWP (Edvardsson Rasmussen et al., 2023a), possibly affecting compliance. Therefore, as cows in the ExtExt treatment were largely selected based on high yield, we expected higher compliance in the ExtExt treatment compared with the previous study, as the herd managers in the current study knew that the cows were expected to be suitable for the treatment. However, complying with a new management routine might be a challenge. Moreover, the effect of having more time to evaluate whether a cow should be inseminated at all might have contributed to the higher proportion of cows not inseminated at all in the ExtExt compared with the ExtConv treatment, as pregnant cows generally run a lower risk of being culled (Gröhn et al., 1998). Another factor affecting compliance in comparison with the previous randomized study (Edvardsson Rasmussen et al., 2023a), where the acceptable VWP was defined as a range, was that in the present study, a maximum VWP of 100 d was set for cows in the ExtConv and ConvConv treatments, meaning that all cows inseminated before 100 DIM were included. In contrast, for the ExtExt treatment all cows inseminated after the 174-d VWP were included. This might mean that cows not showing estrus before 100 DIM were excluded from the ExtConv and ConvConv treatments, which potentially could mean that the fertility results for these treatments might have been slightly overestimated. Regarding the intention to treat analysis for culling rate and disease incidence, this was included to reveal potential bias and detrimental effects due to lack of compliance with the planned VWP. However, this did not change the results which might be explained by the fact that no difference in compliance were observed between the treatments.

The number of cows with high PI or calving problems or disease was small, making it difficult to draw conclusions based on these selection criteria alone. Further, the association between disease and yield is complicated as some cows might be sick due to maladaption to the energy demands of their high milk production (Pascottini et al., 2022), on the other hand clinical disease often result in reduced milk yield (van Soest et al., 2019). In that case, it may be debatable whether cows with calving difficulties or early lactation disease that may risk having a lower milk yield are less suitable for an extended VWP from a milk yield perspective. However, from a fertility perspective, these cows may benefit from a longer time to recover before first insemination (Pascottini et al., 2022). Due to the study design, the fact that the cows in the ConvConv treatment did not have any reported calving problems or disease in early lactation might have affected the comparison to the 2 other treatments. However, because the proportion of sick cows in early lactation in the ExtExt and ExtConv treatments was quite low (10% and 15%, respectively), the effect can be assumed to be limited compared with that of the lower early lactation yield. Moreover, the PI reflects the genetic potential for persistency which in turn only explains part of the phenotypic persistency. Persistency is highly important for successful extension of the VWP; therefore, better prediction of phenotypic persistency is needed and should be investigated in future studies. The aim of the study was to compare different milk production, reproductive, and health traits of cows expected to be suited for extended VWP based on 3 selection criteria. However, the aim was not to evaluate each selection criterion separately, as the study design did not allow for this.

Our results showed that primiparous cows in high-yielding commercial herds with customized VWP, based on the 3 criteria used, had improved fertility and maintained milk yield per day in the calving interval without negative effects on health and culling. Combining the current results with those of previous studies may contribute to improve economic prediction models and support decision making on different management scenarios involving VWP in commercial, high-yielding herds.

CONCLUSIONS

This study showed that by using predefined criteria, it is possible to select primiparous cows suitable for an extended lactation. Our hypothesis could partly be confirmed, as cows suited for and receiving an extended VWP (ExtExt treatment) had better fertility (higher FSCR, fewer NINS, shorter IPL) and higher 305-d and WL yield than cows suited for but randomized not to receive an extended VWP (ExtConv treatment), whereas health, culling, dry period length, and daily yield over the CInt was not affected. A low yield at dry-off is known to be beneficial for udder health, and the ExtExt cows yielded less milk at the last TM before dry-off compared with the ExtConv cows. Our results show that a customized VWP for primiparous cows may be used to increase flexibility in VWP management for herd managers interested in fewer calvings per time period.

NOTES

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Nonstandard abbreviations used: CFI = calving to first service interval; CInt = calving interval; ConvConv = treatment with conventional VWP of ≤90 d; DO = dry-off; ExtConv = treatment with conventional VWP of ≤90 DIM; ExtExt = treatment with extended VWP of ≥185 DIM; FSCR = first service conception rate; HOL = Swedish Holstein; IPL = insemination period length; NINS = number of inseminations per conception; n_{tot} = total number of cows; PI = persistency index; RDC = Red dairy cattle; SNDRS = Swedish National Herd Recording Scheme; TM = test milking; TM3 = third TM; TM8 = eighth TM; TMDO = last TM before dry-off; VWP = voluntary waiting period; WL = whole-lactation; ΔMY = change in MY.

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APPENDIX

Table A1. Description and calculation of included variables (modified after Edvardsson Rasmussen et al., 2023a,b)[1]

Variable	Description/calculation
CInt	Number of days between 2 consecutive calving dates.
Lactation length	The interval between calving and the dry-off date.
DPL	Calving interval minus lactation length.
DPL category	"Short," defined as less than 40 d; "moderate," defined as 40 to 70 d; or "long," defined as more than 70 d.
Milk yield d 4–33	Mean yield d 4–33 for primiparous cows in each herd was calculated. If some daily records were missing, the mean yield was calculated from a regression coefficient derived from daily yields from the first part of the study for the 14 herds that participated in both parts of the study, and for 2 mo mean yield before the start of the study for the 4 new herds. Two herds did not share daily milk yields and for those herds the milk yield and DIM of the first TM was used instead to calculate the mean yield based on the same regression coefficient.
305-d yield	Data on 305-d lactation milk yields retrieved from SNDRS.
Whole-lactation yield	Whole-lactation milk, fat, and protein yields calculated based on test milkings, using the test interval method (Sargent et al., 1968). Dry-off dates reported to SOMRS were used to define end of lactation in these calculations.
ECM yield	Calculated according to the equation by Sjaunja et al. (1990): kg ECM = kg MY × ((38.3 × fat (g/kg) + 24.2 × protein (g/kg) + 783.2)/3,140).
Milk and ECM yield per CInt day	Whole-lactation yield divided by days in the calving interval.
Milk yield at the last TM before dry-off	Daily MY at the TM between 50 and 20 d before dry-off. If the cow had more than 1 TM in this period, data for the TM closest to 50 d before dry-off were used.
Persistency TM3–TM8	Calculated as the change in MY between the eighth TM before DO minus the MY at the third TM, divided by the number of days between these 2 TM, and is expressed as the change in MY per day between the third and eighth TM.
Persistency TM3–DO	Calculated as the change in MY between the last TM before DO minus the MY at TM 3, divided by the number of days between these 2 TM, and is expressed as the change in MY per day between the third test milking and DO.
Total number of inseminations	The number of registered inseminations during the lactation. Double inseminations, defined as 2 inseminations on the same estrus less than 5 d apart, are included.
Inseminations with positive pregnancy diagnosis	The number of inseminations with confirmed positive pregnancy diagnosis performed after the insemination.
Insemination that led to conception	The last insemination in the interval of 280 ± 14 d before the next calving.
Insemination period length	Interval between the first insemination and the insemination that led to conception.
Number of inseminations per conception	Number of inseminations with a positive pregnancy diagnosis divided by the total number of inseminations during the whole first lactation.
First service conception rate	Defined as cows that have had a calf and did not have any insemination before the insemination calculated to be the one that led to calving or cows that had a positive pregnancy diagnosis after the first registered insemination.
Estrus intensity	Estrus intensity reported by the herd manager to SNDRS at the point of first insemination on an ordinal scale from 0 to 5 with 0 representing no signs of estrus and 5 strong signs of estrus, as described by Nyman et al. (2016). Scores of 0 to 2 (representing no or weak estrus signs) were merged due to low frequency of observations, and scores of 4 and 5 were merged because they both represent cows with strong estrus expression and hence have the same biological and practical relevance.
Good udder health	Good udder health was defined as an SCC of <100,000 cells/mL at last TM in the first lactation.
Cow-years at risk	Total number of years that all the cows in each treatment contributed, from calving to next calving, to culling date, or to the date data recording ended (October 22, 2022), whichever occurred first.
Culling rate	Number of cows culled (including mortality) per 100 cow-years at risk during lactation 1.
Disease incidence	Total number of recorded diseases (see also Appendix Table A3) from 33 DIM to next calving, culling date, or the date data recording ended October 22, 2022, whichever occurred first, per 100 cow-years at risk, calculated for each treatment group.

[1]CInt = calving interval; DPL = dry period length; TM = test milking; TM3 = third TM; TM8 = eighth TM; MY = milk yield; DO = dry-off.

Table A2. Recorded diseases and number of culled cows per lactation and 100 cow-years in the study (time at risk) for all cows randomized to "intention to treat" and receiving "per protocol" the planned voluntary waiting period (VWP) treatments (ConvConv, ExtConv, and ExtExt)

	Per protocol			Intention to treat		
Variable	ExtExt (n = 120)	ExtConv (n = 139)	ConvConv (n = 132)	ExtExt (n = 167)	ExtConv (n = 169)	ConvConv (n = 177)
Disease cases from 33 DIM	20	31	14	24	38	16
Culled cows	16	21	16	35	35	47
Cow-years in study	159	141	132	205	173	167

Table A3. Number of disease registrations per diagnosis category from 33 DIM during lactation 1 for each VWP treatment group

	Intention to treat		
Diagnosis	ExtExt (n = 167)	ExtConv (n = 169)	ConvConv (n = 177)
Subclinical mastitis	9	15	6
Mastitis	5	8	6
Other	3	7	3
Leg/hoof lesion	3	4	0
Reproductive disease	2	2	1
Accident/trauma	1	1	0
Metabolic	1	1	0
Total number of diagnoses	24	38	16