



Extended lactations in multiparous dairy cows

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Licentiate Thesis
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SWEDISH UNIVERSITY
OF AGRICULTURAL
SCIENCES

Licentiate Thesis

Uppsala 2025

Rapporter från institutionen för tillämpad husdjursvetenskap och välfärd

2025:12

Cover: Cow and calf. Maria Nyberg, Växa 2025

ISSN (print version) 2004-9803

ISSN (electronic version) 2004-934X

ISBN (print version) 978-91-8124-023-8

ISBN (electronic version) 978-91-8124-024-5

<https://doi.org/10.54612/a.72ch2hvv0q>

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Print: SLU Grafisk service, Uppsala 2025

Extended lactations in multiparous dairy cows

Abstract

The substantial increase in milk yield capacity in modern dairy herds has generated interest in extending voluntary waiting period (VWP) as a strategy to prolong the calving interval (CInt). This Licentiate thesis investigated the effects of extended VWP on fertility, milk production, and culling in second-parity cows, as well as associations between CInt, milk yield in second parity and mortality during the first month after the third calving. In a randomized controlled study of 819 second-parity cows from 12 high-yielding commercial herds, cows were allocated to either a conventional (50-day) or extended (140-day) VWP. The VWP treatments resulted in calving intervals of 12.5 and 14.0 months, respectively, but had no effect on daily milk yield per calving interval day, milk yield per lactating day, or dry period length. Fertility improved in the extended VWP group, as indicated by a higher pregnancy proportion in the first service, fewer inseminations per calf and a shorter insemination period. No effect on culling rate was observed. In a retrospective cohort study, data from the Swedish Official Milk Recording Scheme was analyzed to evaluate associations between CInt and on-farm mortality after the third calving. An increased risk of mortality was detected in cows with CInt longer than 13 months. Mortality risk was also associated with high milk yield, both as a 305-day yield and as the yield at the last insemination relative to the herd average at the third test milking. In the subset of the most high-yielding herds, this difference was not statistically significant. Together, these findings indicate that an extended VWP can improve fertility in second-parity cows without compromising milk production or affecting culling rates. However, long CInt were associated with an increased risk of on-farm mortality within 30 days after the subsequent calving.

Keywords: Lactation length, extended calving interval, reproductive management, animal welfare, dairy cow mortality

Förlängd laktation för omkalvande mjölkkor

Abstract

Mjölkkornas ökade avkastningskapacitet i dagens mjölkbesättningar har medfört ett ökat intresse för förlängd frivillig väntetid (FVT) som en strategi för att förlänga kalvningsintervallet. I denna licentiatuppsats undersöktes effekterna av förlängd FVT på fertilitet, mjölkproduktion och utslagning i andra laktationen, samt samband mellan kalvningsintervall, mjölkavkastning i andra laktationen och kodödlighet under första månaden efter tredje kalvningen. I en randomiserad kontrollerad studie med 819 kor i andra laktationen från 12 högavkastande besättningar tilldelades korna antingen en konventionell (50 dagar) eller förlängd (140 dagar) FVT. Behandlingarna resulterade i kalvningsintervall på 12,5 respektive 14,0 månader, utan att mjölkavkastning per dag mellan kalvningarna, mjölkavkastning per laktationsdag eller sinperiodens längd skilde sig åt som en effekt av behandling. Fertiliteten var bättre i gruppen med förlängd FVT, vilket visades genom en högre dräktighetsprocent vid första semineringen, färre semineringar per född kalv samt en kortare semineringsperiod. Ingen effekt på utslagningsfrekvens observerades. I en retrospektiv kohortstudie analyserades data från Kokontrollen för att utvärdera samband mellan kalvningsintervall och dödlighet inom 30 dagar efter tredje kalvningen. En ökad risk för dödlighet påvisades bland kor med ett kalvningsintervall längre än 13 månader. Risken var också associerad med hög mjölkavkastning, både som 305-dagarsavkastning och som avkastning vid sista semineringen. I de mest högavkastande besättningarna var skillnaden dock inte statistiskt säkerställd. Sammantaget visar resultaten att en förlängd FVT kan förbättra fertiliteten hos kor i andra laktationen utan att påverka mjölkproduktion eller utslagningsfrekvens. Däremot var långa kalvningsintervall kopplade till en ökad risk för dödlighet inom 30 dagar efter påföljande kalvning.

Nyckelord: Laktationslängd, förlängt kalvningsintervall, reproduktionsstyrning, djurvelfärd, kodödlighet

Preface

Inchaisri et al. (2011) noted, “Determining the optimal VWP from field data is difficult and unlikely to happen”, highlighting the complexity of implementing controlled treatments under real-world conditions.

Dedication

♡ To my family - tumbling along on this thesis ride.

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List of publications

This thesis is based on the work contained in the following papers, referred to by Roman numerals in the text:

- I. Hansson, A., Holtenius, K., Båge, R., Lindberg, M., Kronqvist, C. (2025). Effect of voluntary waiting period length on milk yield, fertility and culling in high yielding, second-parity cows. *Journal of Dairy Science*, (accepted)
- II. Hansson, A., Lindberg, M., Båge, R., Kronqvist, C. An observational retrospective study on the association between calving interval and mortality among Swedish dairy cows. (manuscript)

Paper I are reproduced with the kind permission of the *Journal of Dairy Science*.

The contribution of Annica Hansson to the papers included in this thesis was as follows:

- I. Participated in planning the study in collaboration with the supervisors and had the main responsibility for coordinating and running the trial with their support. The results were analyzed in collaboration with the statistical unit and supervisors. Preliminary findings were presented at two conferences with support from the supervisors. Drafted and revised the manuscript in collaboration with the co-authors and, under supervision, managed correspondence with the journal and revisions during the review process.
- II. Participating in planning the study in collaboration with the supervisors. Involved in creating the statistical models. Drafted the manuscript.

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Abbreviations

CFI	Calving to first insemination interval
CInt	Calving interval
DIM	Days in milk
DPL	Dry period length
ECM	Energy-corrected milk
FSCR	First service conception rate
FSP	First service pregnancy
IPL	Insemination period length
MY	Milk yield
NINS	Number of inseminations per cow
SOMRS	Swedish official milk recording scheme
VWP	Voluntary waiting period

1. Introduction

Dairy cattle (*Bos taurus*) were domesticated about 10,500 years ago and have since diversified into more than 1,000 breeds, primarily used for meat and milk production (and, in some regions, also for draught power). In both their natural and extensively managed state, cattle typically reproduce on an annual cycle, giving birth once a year in synchrony with pasture availability. This natural reproductive rhythm corresponds to the 12-month calving interval that would later become the reference point for modern dairy production.

In pasture-based dairy production systems, spring calving aligns with the grazing season to optimize milk production. In contrast, intensive systems that provide high-quality forage and concentrates year-round benefit from evenly distributed calving to maximize resource use. In such a system, a 12-month calving interval (**CInt**) is often recommended and has long been considered economically optimal (Schneider et al., 1981; Strandberg and Oltenacu, 1989; Steeneveld and Hogeveen, 2012). To achieve this interval, the voluntary waiting period (**VWP**) is typically set to about 50 days after calving, after which cows are inseminated at the first observed estrus (Rehn et al., 2000).

As milk yields have increased in modern dairy production, the traditional 12-month CInt has been questioned, and several studies have proposed the use of extended intervals (Arbel et al., 2001; Österman and Bertilsson, 2003; Niozas et al., 2019a).

However, longer calving intervals have been identified as one factor associated with an elevated mortality risk (Raboisson et al., 2011; Alvåsen et al., 2012; Alvåsen et al., 2014a). In recent decades, on-farm mortality (including both euthanasia and death) has increased in several countries with intensive dairy production, such as Sweden (Alvåsen et al., 2012), Denmark (Thomsen et al., 2004) and the United States (Miller et al., 2008). This trend has been linked to broader structural changes in the dairy sector, including economic pressure towards farm consolidation, increased productivity and more intensive production systems, (McConnel et al., 2008).

High mortality rates are not only economically burdensome but also emotional distressing for dairy producers. Furthermore, public concern around animal welfare, of which mortality rates are a key indicator (de

Graaf et al., 2016), has intensified. This underscores the importance of clarifying whether extended calving intervals contribute to increased mortality, in order to avoid unintended negative consequences when evaluating longer intervals in today's high-yielding dairy herds.

2. Background

2.1 Dairy production

2.1.1 Swedish dairy statistics on milk yield, fertility and mortality

Swedish dairy production has undergone substantial structural changes over recent decades, marked by a shift toward fewer but larger herds. While the total number of dairy cows has steadily declined, milk production per cow has increased markedly. Statistics from the Swedish official milk-recording scheme (**SOMRS**), which collects data on milk yield (**MY**), fertility and health from affiliated herds, illustrates this development. In the early 1980s, the average Swedish dairy cow produced approximately 6,000 kg energy-corrected milk (**ECM**) annually. During the most recent milk-recording year (September 2023 to August 2024), the 1,670 herds enrolled in SOMRS, comprising 173,318 cows or 73.6% of the dairy cows in Sweden, reported an average production of 11,224 kg ECM per cow and year (Växa, 2025).

Fertility data from the same period shows that, on average, cows were first inseminated 83 days after calving, required 1.8 inseminations, and had a mean CInt of 13.0 months. Variation at the herd level was substantial; the 10% of herds with the shortest CInt averaged less than 12.2 months, whereas the 10% with the longest CInt exceeded 15.2 months.

In Sweden, fertility management is primarily based on detection of spontaneous estrus, with approximately 2% of milk-recorded cows receiving a series of inseminations following treatment for fertility disorders. Insemination practices have also shifted over time, with an increasing proportion now performed by farmers themselves; in 2024, 83% were carried out by farmers (Växa, 2025).

On-farm mortality, defined as euthanasia or unassisted death, is presented in SOMRS as the number of deaths per year per 100 cows at the herd level. Alvåsen et al. (2012) examined cow mortality in Sweden and reported an increasing trend over time. In Sweden, the median herd mortality rate was 4.7 % (Växa, 2025).

2.1.2 Lactation yield measurements

The 305-day MY represents the total amount of milk produced from calving until 305 days postpartum and aligns with a conventional 12-month CInt, allowing for comparisons that account for a ten-month lactation and two-month dry period length (**DPL**). However, when lactation length is shorter than 305 days, due to a short CInt with a maintained two-month dry period or early dry-off for health reasons, non-lactating days are still included in the measure. Conversely, the 305-day MY can be biased upward in cows with delayed pregnancy, since pregnancy typically reduces daily MY (Strandberg and Lundberg, 1991; Andersen et al., 2011; Chen et al., 2024). Total lactation yield reflects the full milk output until dry-off, regardless of lactation length. Thus, the 305-day MY is strongly affected by CInt and therefore less suited to the comparison of cows or herds with different CInt strategies.

Milk yield per day of calving interval has been proposed as a useful performance measure to better evaluate lactations of varying length (Österman and Bertilsson, 2003; Lehmann et al., 2016). Kok et al. (2016) compared three standardized measures, all expressed in kg ECM/day, for cows with different DPL: the conventional 305-day yield, a 365-day yield calculated by adding milk produced in the 60 days before calving to the 305-day yield, and an effective lactation yield defined as milk produced from 60 days before one calving to 60 days before the next. The effective lactation yield accounted for variations in both DPL and CInt.

2.1.3 Lactation curve patterns in primiparous and multiparous cows

Lactation persistency, defined as the ability to maintain production after peak, is a key factor when aiming for lactations beyond the standard 10-month duration (Dekkes et al., 1998). Peak yield, the highest daily milk production during a lactation, typically occurs around two months postpartum in high-yielding herds (Lehmann et al., 2017). Primiparous cows generally show higher lactation persistency than multiparous cows and have therefore often been identified as the most suitable candidates for extended lactations (Rehn et al., 2000; Lehmann et al., 2019). Multiparous cows, however, usually achieve higher peak yields than primiparous cows (Burgers et al., 2021b), which may partly offset their lower lactation persistency. This suggests that under certain conditions, such as when milk yield at dry-off is excessively high, multiparous cows may benefit from

extended lactations, potentially improving both cow welfare and overall production efficiency.

2.1.4 Voluntary waiting period and calving-to-first-insemination

The voluntary waiting period refers to the interval between calving and when farmers intend to rebreed a cow, and its outcome is reflected in the herd-average calving-to-first-insemination (CFI). Theoretically, if all cows were cyclic and all estrus were detected and inseminated, the deviation between VWP and CFI would equal half an estrus cycle. In practice, deviations originate from involuntary or voluntary causes. Involuntary reasons include individual variation in postpartum recovery and resumption of estrus (Ratnayake et al., 1998; Lopez et al., 2004), as well as management-related factors such as inconsistent adherence to VWP protocols or limited estrus detection. The latter may be due to inadequate observation routines or insufficient technological support. Extended VWP, in contrast, reflects deliberate management adjustments, as shown in the commercial herds where farmers extend VWP in response to factors such as high daily MY or calf surplus (Lehmann et al., 2016; Burgers et al., 2021b).

2.1.5 Submission and conception rate

The submission rate reflects the efficiency of estrus detection and thereby strongly influences calving interval length. Submission rate is defined as the proportion of eligible cows, those beyond the voluntary waiting period, that are inseminated within a 21-day period. In Swedish herds, reported submission rates vary widely, from as low as 20% to over 90%. High rates are generally associated with effective estrus detection routines, the use of reliable sensor technologies, balanced nutrition, non-slippery flooring, and cows in overall good condition.

The conception rate measures the proportion of insemination that result in confirmed pregnancy. In SOMRS, average herd-level conception rates generally range from 45% to 50%, although substantial variation exists.

2.1.6 Dry period

The dry period length refers to the interval between the last milking of one lactation and the subsequent calving. Farmers are generally advised to aim

for a DPL of 6 to 8 weeks, as this duration is considered optimal for udder recovery and maximizing milk production in the following lactation (Dias and Allaire, 1982; Koyama et al., 2024).

High daily MY prior to dry-off has been associated with an increased risk of udder health problems at the subsequent calving (Rajala-Schultz et al., 2005). In addition, sudden dry-off in high-yielding cows can cause udder pressure and pain, leading to stress and raising important animal welfare concerns regarding dry-off strategies (Bertulat et al., 2013). Several studies have been conducted to identify best practices and provide evidence-based recommendations for dry-off management (Odensten, 2006; Andrée O'Hara et al., 2018).

An extended CInt can lead to lower MY at dry-off (Niozas et al., 2019a), which may reduce the risk of mastitis during transition periods (Rajala-Schultz et al., 2005). However, if lactation length does not increase proportionally with the CInt, the dry period may exceed the recommended 6–8 weeks. Studies on extended VWP have shown that longer VWPs are associated with longer DPL (Rehn et al., 2000; Edvardsson Rasmussen et al., 2023). Andrée O'Hara et al. (2020) reported that a DPL exceeding 70 days was associated with lower milk production, reduced fertility, and increased risk of culling in the following lactation. Extended CInt may also increase the risk of excessive weight gain during the extended period of low production (Burgers et al., 2021a). Over-conditioning, in turn, may elevate the risk of complications in the subsequent calving.

2.1.7 Calving interval

The calving interval is defined as the time between two consecutive calvings. It consists of the period from calving to conception, denoted as days open, and the duration of gestation, which in dairy cows is approximately 280 days. Thus, the cow must conceive around three months postpartum to maintain a 12-month CInt. This means that a 13.0-month average CInt indicates that, on average, cows conceived around 115 days after calving (SOMRS, 2025).

With increasing MY capacity in modern dairy cows, there has been growing interest in re-evaluating the optimal length of the CInt (Österman, 2003; Niozas et al., 2019a; Römer et al., 2020). In Sweden, calculations have estimated an economic loss of approximately 13 SEK per day from involuntarily prolonged CInt beyond 12 months (Engelbrekts, 2015). This

estimate was based on the assumption that the extended portion of lactation was associated with reduced daily MY and, consequently, lower milk income.

However, this may be an overly simplified way to estimate the economic impact of varying CInt lengths. The negative effect on milk production from delayed conception has shown to be minimal for cows with more persistent lactations, a trait more common in primiparous cows, and for higher-parity cows when delayed conception occurs early in lactation, whereas delays later in lactation tended to result in greater losses (Steenefeld and Hogeveen, 2012; Burgers et al., 2021a).

2.2 Studying voluntary waiting period and calving interval

2.2.1 Scientific method, random controlled study vs retrospective study

Randomized controlled trials, considered the gold standard for establishing cause-and-effect relationships, generate primary data and allow prospective evaluation of defined interventions. However, they are often constrained by costs, time requirements, limited herd numbers, and challenges with compliance in commercial settings (Tripepi et al., 2020; Edvardsson Rasmussen et al., 2023)

In contrast, retrospective studies rely on pre-existing secondary data to explore associations between factors and outcomes. They are often based on large datasets, making them particularly useful for studying rare events or long-term patterns under commercial conditions. However, they may be limited by data quality, unmeasured confounding and an inability to establish causality (Talari and Goyal, 2020).

2.2.2 Milk yield variables

Total MY (Österman and Bertilsson, 2003; Edvardsson Rasmussen et al., 2023) and 305-day yield (Burgers et al., 2021b; Edvardsson Rasmussen et al., 2023) have been found to increase with CInt. To compare cows with different CInt lengths, milk yield expressed per day of the calving interval or per lactating day provides the most appropriate measure. Among multiparous cows, the effect of extended VWP on yield is less consistent.

Some studies reported lower average daily yield per CInt day (Arbel et al., 2001; Österman and Bertilsson, 2003; Burgers et al., 2021a). while others found no effect (Rehn et al., 2000; Stangaferro et al., 2018). One study even reported increased yield per CInt day, but as it also evaluated the effect of bST, the results are not comparable (Van Amburgh et al., 1997).

Differences between studies may be explained by variation in lactation curves or DPL. However, DPL did not differ between groups in the study by Arbel et al. (2001). In the study by Österman and Bertilsson (2003), DPL differed among cows milked twice daily but not among those milked three times daily.

Retrospective data has also been used to study customized VWP, where farmers selectively extend the interval for individual cows based on specific criteria (Lehmann et al., 2016; Lehmann et al., 2017). Analysis of four Danish herds practicing extended VWP, often motivated by high milk yield at dry-off or fertility concerns, showed that cows maintained similar milk production per feeding day for calving intervals up to 19 months. However, daily milk yields close to dry-off declined when calving intervals exceeded 15 months in second-parity cows and 13 months in older cows. Dry period length also increased by 3 to 5 days with extended calving intervals (Lehmann et al., 2016).

2.2.3 Fertility variables

A randomized controlled study at the Swedish University of Agricultural Sciences was conducted in two research herds, with VWP set to 50 and 140 days in one herd and 50 and 230 days in the other herd. In both herds, inseminations were allowed within a 130-day window, with a maximum of five inseminations per cow. Bertilsson et al. (1997) reported results from the first lactation and found no significant differences in first-service conception rate (FSCR) or pregnancy rate. However, cows in the VWP 50 group required significantly more anestrus treatments than those in the VWP 140, a finding later confirmed by Niozas et al. (2019b) and Němečková et al. (2015). Estrus intensity increased until about 80 days into milking and then stabilized, a pattern also observed in commercial herds (Edvardsson Rasmussen et al., 2023). Subsequent analysis comparing VWP 50 and 140 days showed that extended VWP was associated with higher FSCR, a shorter interval from first service to conception and fewer inseminations overall (Larsson and Berglund, 2000).

Retrospective data has been used to study customized VWP, where farmers selectively extended the interval for individual cows based on specific criteria (Lehmann et al., 2016; Lehmann et al., 2017; Burgers et al., 2021b).

Burgers et al. (2021b) found that a longer CInt was often not planned as the majority of cows had been inseminated earlier but failed to conceive. Also, cows in the shortest VWP group had longer than expected CInt, primarily due to a higher number of inseminations (NINS) required to achieve pregnancy.

2.2.4 Culling

Culling has been evaluated in some studies on VWP with varying results. Arbel et al. (2001) and Rasmussen et al. (2023) reported no significant differences in culling rates between treatment groups. Larsson and Berglund (2000) found that more cows were culled due to poor fertility, defined as a failure to conceive after five inseminations within 130 days, in the VWP 50 group. Niozas et al. (2019b) observed a tendency towards higher culling rates in cows assigned to a VWP of 180 days compared to those with VWP of 40 days.

2.3 On-farm cow mortality

2.3.1 On-farm cow mortality

On-farm cow mortality in Swedish dairy herds has increased over time, rising from 5.1 to 6.6 events per 100 cow-years between 2002 and 2010 (Alvåsen et al., 2012). This trend was partly attributed to the ongoing intensification of dairy production, characterized by larger herd sizes, a shift from tie-stall to free-stall housing systems, and an increasing proportion of Holstein cows relative to Swedish Red cows as risk factors. The distribution of on-farm cow mortality shows that it frequently occurs close to calving (Dechow and Goodling, 2008; Raboisson et al., 2011; Alvåsen et al., 2014a). Thomsen (2023) further reported that approximately 32% of deaths occurred within 30 days in lactation and 49% within 90 days. With increasing parity, a larger proportion of the deaths occurred early in lactation; for example, in the fifth parity, 25% of all deaths occurred within the first 8 days of lactation.

2.3.2 On-farm cow mortality and calving interval

On-farm mortality has been shown to increase with extended CInts. In a study of 6,898 Swedish herds between 2002 and 2010, Alvåsen et al. (2012) confirmed an overall upward trend in mortality and found that herds with predominantly HOL cows had higher rates than those with SR. Herds with an average CInt shorter than 12.8 months had a mortality rate of 5.5% per cow-year, compared with 7.4% in herds with a CInt longer than 13.8 months. Supporting these findings, Raboisson et al. (2011), also observed a positive association between longer CInt and mortality in French dairy herds in 2005–2006. They further suggested that farmers who maintained shorter CInt often concentrated calvings in autumn to align with favorable seasonal conditions, reflecting a higher overall level of management that may reduce mortality risk.

At the cow-level, Alvåsen et al. (2014a) followed cows from calving to the next calving and found that longer calving intervals were associated with the significantly increased danger of on-farm mortality. Calving intervals were categorized as <11.7 months, 11.7–14.2 months, and >14.2 months, with mortality risk rising across the groups. The authors suggested that CInt may serve as a proxy for management quality, as farmers achieving shorter intervals were considered to be more likely to prevent problems that could lead to cow death.

Longer calving intervals have also been linked to higher mortality in U.S. dairy herds. Shahid et al. (2015) analyzed both herd and cow-level data from the Midwest between 2006 and 2010 and found that, at the herd level, compared with a reference of 12–14 months, herds with a CInt of 15–16 months had a 16% higher mortality risk, and herds with >16 months had a 21% higher risk. The results agreed with previous studies (Raboisson et al., 2011; Alvåsen et al., 2012) that shorter herd-level CInt may serve as a proxy for better overall management, which in turn reduces mortality. At the cow level, longer CInt was also associated with higher mortality risk, and the authors speculated that over-conditioning during extended lactations could be one contributing factor.

2.3.3 Mortality rate and its association to milk yield

At the cow-level, higher MY in the previous lactation has been associated with increased on-farm mortality risks (Alvåsen et al., 2014a). Conversely, low yield or missing values at the first test-milking have also been linked

to elevated mortality risk (Alvåsen et al., 2014a). At the herd-level, however, several studies report the opposite pattern, with high-producing herds showing lower mortality risk than lower-producing herds (Thomsen et al., 2006; Burow et al., 2011; Alvåsen et al., 2012). These findings indicate that the association between milk yield and mortality differs between the individual and herd levels, reflecting both biological and management factors.

2.4 Description of the knowledge gap

Extended VWP has been associated with both advantages and drawbacks in MY. Persistency patterns and parity influence the outcome, with primiparous cows often identified as the most suitable candidates, while multiparous cows may also benefit under specific conditions, such as high MY at dry-off. The effect of extended VWP on fertility has been evaluated in both research- and commercial herds. Randomized studies indicate potential benefits, such as higher FSCR and fewer inseminations per pregnancy, while other studies report neutral outcomes. However, research herds often operate under controlled protocols with fixed limits for insemination attempts, conditions that may not be feasible in commercial settings. Culling, meanwhile, is a multifactorial outcome, influenced by both biological and management decisions, and results vary depending on study design and how culling is defined.

A recent study in Swedish commercial herds by Rasmussen et al. (2023) provided valuable evidence by evaluating extended VWP strategies in primiparous cows. However, controlled studies focusing on cows in their second lactation, which have higher milk yield capacities, remain rare. Thus, applying the same interventions and outcome variables to this group has the potential to generate new knowledge and complement existing findings from primiparous cows.

At both the herd and cow levels, longer calving intervals have been associated with higher on-farm mortality. Studies consistently confirm this association, although interpretations differ. Shorter calving intervals may serve as a proxy for better management, although cow-level mechanisms such as over-conditioning during extended lactations have also been proposed. Despite this consistency, the causal pathways linking extended calving intervals, milk yield and mortality remain poorly understood.

3. Aims of the thesis

The overall aim of this thesis was to investigate the effect of extended VWP for high yielding dairy cows in their second lactation in commercial herds. The specific objectives were:

- To investigate the effect of extended VWP on milk production, fertility and culling in second-lactation cows within high-yielding commercial dairy herds through a randomized controlled study,
- To investigate the relation between CInt during the second lactation on the risk of on-farm mortality within 30 days after the subsequent calving, and to examine the association with MY during second lactation and daily MY at the time of conception.

4. Material and methods

A randomized controlled trial, considered to be the gold standard for establishing cause-and-effect relationships, was conducted to investigate the effects of different VWP on fertility and milk yield in second lactation cows. In paper I, cows in 12 commercial herds were randomly assigned to either a 50-day or a 140-day VWP.

A retrospective cohort study was conducted to investigate the association between calving interval (CInt) and on-farm mortality. In Paper II, a dataset from the Swedish Official Milk Recording Scheme (SOMRS), spanning six years, was used to examine mortality within 30 days after calving in relation to CInt. For detailed description, the reader is referred to the individual papers.

4.1 The randomized controlled trial

4.1.1 Study design and herd recruitment

Approximately 50 commercial dairy herds in southern Sweden were initially contacted and invited to participate in a study on optimal calving interval, referred to here as Paper I. To be eligible for the study, herds had to be enrolled in the SOMRS, consist of more than 100 cows and have an annual average MY above the national average of 10,400 kg ECM. The participating herds varied in size, feeding strategies, milkings per day (either two or three), milking system (parlour or automatic) and breed composition, and included both stable and expanding operations. This variation in herd characteristics reflects the diversity of commercial dairy production systems. The study aimed to randomize second-lactation cows to either a VWP of 50 or 140 days, based on their ear tag number (even or odd). Once 14 herds had agreed to participate, it was estimated that at least 500 cows would complete a full lactation, calving for a third time, meeting the target sample size to evaluate outcomes related to milk production, fertility, and culling.

Herd enrolment began with cows calving their second calf in October 2019. Allocation continued for cows calving in the following year. A dedicated Facebook group was created as a communication platform, offering information and allowing participants to share experiences. For

example, best practice for maintaining compliance with the protocol was discussed within the group. Participation was voluntary, and herds could withdraw from the study at any time. One herd was excluded early in the process after leaving SOMRS, and two new herds were recruited as replacements. By December 2020, a total of 992 cows had calved for the second time and been enrolled in the study, concluding the enrolment phase.

To allow sufficient time for cows to be inseminated, conceive, and calve for a third time, enabling evaluation of fertility and milk production, the study period continued until June 2022. Throughout this period, participating herds received regular updates with preliminary results.

Protocol adherence and herd selection

Three herds were excluded from the study due to insufficient adherence to the treatment protocol, as the mean CFI difference between the two treatment groups in these herds was less than 28 days (5, 18 and 26 days). In the refined dataset, which included 12 herds, the mean CFI in the VWP50 group decreased slightly from 72.2 to 69.9 days, while the VWP140 group increased from 121.0 to 124.0 days. The standard deviation also decreased by more than 2 days in both groups, indicating improved consistency in protocol implementation.

4.1.2 Data collection and processing

Cow-level data was retrieved from SOMRS. Variables included in the analysis were breed, MY, fat and protein percentages, and dates for inseminations, calvings, dry-off and culling (including recorded reason for culling).

Total MY during the second lactation and MY during the first 100 days of the third lactation were calculated using monthly test-milking data from SOMRS. The average daily MY was calculated for the two periods per lactating day and per calving interval day. All yield values were expressed in kg ECM.

Fertility was evaluated separately for cows that calved a third time and those that did not. Pregnancy was defined as an insemination followed by a third calving. Outcome variables included first service pregnancy (**FSP**), referring to pregnancies resulting from a single insemination. Additionally, the number of inseminations per cow (**NINS**) was recorded, along with the

insemination period length (**IPL**), defined as the number of days from the first to the last insemination.

Culling was evaluated based on three proportions, the total proportion of culled cows, the proportion of cows not inseminated, and the proportion culled due to reduced fertility relative to all cows. On-farm mortality, including unassisted death and euthanasia, was included in culling.

4.1.3 Statistical analysis

In the field study evaluating the effect of VWP, linear mixed models were used to analyse continuous outcome, including CFI, IPL, DPL, CInt and various milk yield measures. The count variable NINS was analysed using a Poisson regression model. Binary outcomes, such as insemination status, FSP, overall pregnancy status and culling, were analysed using a logistic regression. A 95% confidence level was applied in all analyses.

Fixed effects included VWP treatment group (two levels), breed (three levels) and their interaction. Farm (12 levels) was included as a random factor to account for variations at the herd level.

Further details on the statistical models are provided in Paper I.

4.2 The retrospective study

4.2.1 Data retrieval and study population

This analysis, referred to as Paper II, aimed to explore associations between CInt, MY, and on-farm mortality using a retrospective study design. Data was retrieved from the SOMRS, and the study covered second lactations between 2015 to 2022. Herds with fewer than 30 cows were excluded. Breeds were classified in four groups: SR, HOL, crosses between these breeds and crosses with other breeds. The outcome variable, on-farm mortality, was defined as slaughter on the farm, euthanasia on the farm or unassisted death occurring within 30 days after the third calving.

In total, 147,088 cows with recorded second and third calvings were identified. To be included in the analysis, cows were required to have a complete 305-day milk yield record, sufficient test-day data, a full-term pregnancy, and a DPL of no more than 135 days. After applying these criteria, 139,444 cows remained in the dataset. For analysis, CInts were

categorized into four groups: less than 356 days (CIS), 356–395 days (CIM), 396–456 days (CIExt), and more than 456 days (CIL).

To test whether high milk production influenced the association between CInt and mortality, 305-day ECM yield was used as an indicator of production level. Cows were categorized into four MY classes: $\leq 9,000$ kg ECM (MY1), 9,001–11,000 kg ECM (MY2), 11,001–12,000 kg ECM (MY3) and $\geq 12,001$ kg ECM (MY4).

A second test aimed to test the potential risk of over-conditioning during the lactating phase after conception. Cows were classified based on their MY at test milking closest to insemination. This yield was compared to the herd average for the third test milking within the same fiscal year. Three categories were defined: >2 kg above herd average, within ± 2 kg of the herd average and >2 kg below the herd average.

To analyse mortality in relation to CInt under high-performing conditions, a subset of well-managed herds was selected based on average annual MY in fiscal years 2016 and 2022. Herds ranking among the top 300 in both years were classified as well-managed ($n = 150$). A total of 18,115 cows from these herds were included in the analysis. The model evaluating the effect of MY at the time of insemination was applied to this subset of cows from high-yielding herds.

4.2.2 Statistical analysis

The associations between CInt group and yield class were evaluated using a Chi2-test. The effect of CInt and MY on on-farm mortality was assessed using a logistic regression with a logit link, modelling the probability of mortality within 30 days postpartum.

All cows were tested for the effect of 305-day MY, and MY at the test day closest to the date of conception. In these models, fixed effects included breed (four levels), calving interval (CInt; four levels), 305-day MY (four levels) or energy-corrected milk (ECM) at last insemination (three levels), and the interaction between CInt and yield class. Herd was included as a random effect.

In the model limited to cows from well-managed, high-yielding herds, fixed effects included CInt (four levels), breed (four levels), 305-day MY (three levels), and the interaction between CInt and MY. Herd (150 levels) was again included as a random effect.

A 95% confidence level was applied for all statistical analyses. Further details on the statistical models are provided in Paper II.

5. Main findings

5.1 Effect of allocation to different voluntary waiting periods

Both the CFI and the CInt were shorter in the VWP50 group (68 and 381 days) than in the VWP140 group (127 and 427 days). The distribution for CFI is given in Figure 1. However, DPL did not differ significantly between the groups (64 vs. 67 days).

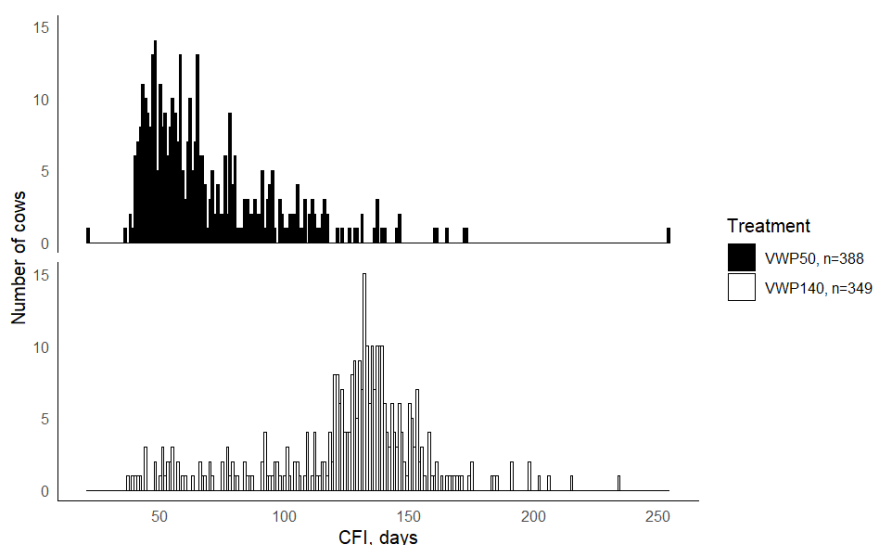


Figure 1. Day from calving to first insemination (CFI) for cows assigned to either a voluntary waiting period (VWP) of 50 days or a voluntary waiting period of 140 days. From Paper I.

5.1.1 Milk production

The longer lactation period in the VWP140 group contributed to a higher total MY compared to the VWP50 group (14,623 vs. 12,881 kg ECM), as well as a higher 305-day MY (12,785 vs 12,064 kg ECM). However, there were no significant differences between groups in kg ECM per day of CInt (34 kg) or ECM per day of lactation (40.5 kg). Daily MY, expressed as ECM, close to dry-off was lower in the VWP140 group (32.5 kg) compared

to the VWP50 group (33.7 kg). Milk production during the first 100 days of the third lactation did not differ between groups (4,650 kg ECM).

5.1.2 Fertility traits

Extending the VWP to 140 days, compared to the conventional 50-day VWP, improved fertility parameters. First service pregnancy rate was significantly higher in the VWP140 group than in the VWP50 group (62% vs. 46%), as indicated in Figure 2. The number of inseminations per calving was lower in the VWP140 group compared to the VWP50 group (1.61 vs. 2.02), and the IPL was shorter (20.8 vs. 35.6 days). This is illustrated as a steeper probability-curve in Figure 3, indicating improved insemination efficiency.

As expected, based on the study design, the interval from calving to first insemination was longer in the VWP140 group (127 vs. 68 days), resulting in a longer CInt (427 vs. 381 days). Notably, a 60-day extension of CFI led to only a 46-day extension of the CInt.

5.1.3 Culling

In total, 425 cows were allocated to the VWP50 group, of which 105 (24.7%) were culled, including 37 cows (8.7%) that were culled without having been inseminated. In the VWP140 group, 394 cows were allocated, of which 120 (30.5%) were culled, including 45 cows (11.4%) without insemination.

Reduced fertility was reported by farmers as the reason for culling in 17 cows (4%) in the VWP50 group and 24 cows (6.1%) in the VWP140 group. However, treatment group did not significantly affect the risk of being inseminated nor the risk of being culled during the second lactation.

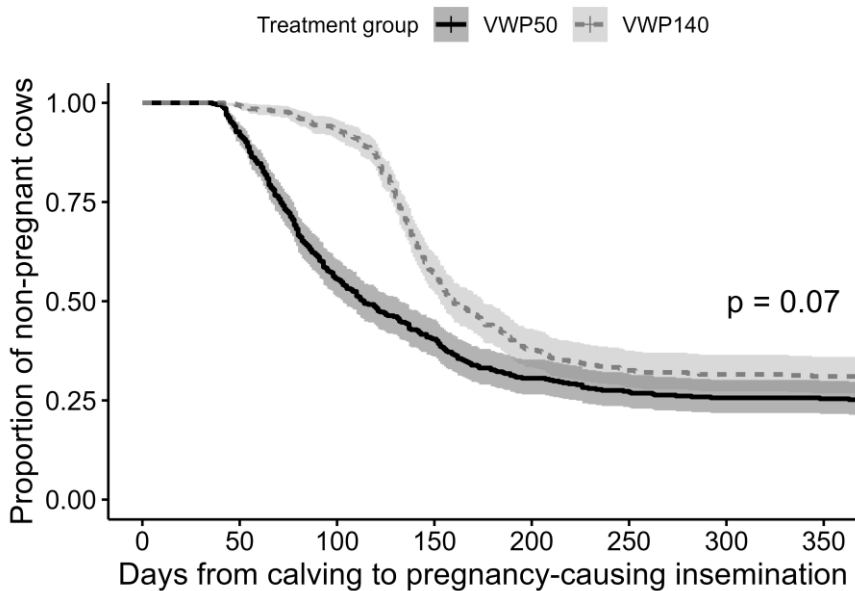


Figure 2. A Kaplan-Meier survival curve, showing the proportion of non-pregnant cows over time (days from calving to pregnancy-causing insemination) for the two treatment groups: VWP50 (solid black line) and VWP140 (dashed grey line). The extended voluntary waiting period (VWP140) resulted in a delayed onset of insemination, a shorter insemination period and a tendency toward a lower proportion of cows becoming pregnant compared with VWP of 50 days ($p=0.007$). Shaded areas represent 95% confidence intervals.

5.2 On-farm mortality and calving interval

5.2.1 Results from the full dataset

Of the 139,444 cows that calved for the third time, 1,482 (1.78) died or were euthanized within the first 30 days of the third lactation.

Among cows with the shortest CInt, 35% belonged to the lowest yield class, whereas in the longest CInt group, 65% were classified as low-yielding. Likewise, the proportion of high-yielding cows declined with increasing CInt, from 36% in the shortest group to only 14% in the longest.

A calving interval longer than 13 months was again associated with an increased risk of mortality within 30 days postpartum. In addition,

mortality was higher among cows with a high MY at the time of insemination compared to those with average or low yield.

No interaction effects were observed between CInt and any of the yield classification variables on mortality within 30 days after third calving.

In the model where yield was classified based on 305-day MY, the probability of early-lactation on-farm mortality increased among cows with a CInt longer than 13 months and was further elevated for those exceeding 15 months, regardless of yield class. Additionally, cows with a 305-day yield above 12,000 kg ECM had a higher probability of on-farm mortality compared to cows producing less than 9,000 kg ECM.

In the second model, where yield was defined as daily ECM at the last insemination relative to the herd average at the third test milking, the probability of on-farm mortality also increased with CInt over 13 months, and further with CInt exceeding 15 months, independent of yield class. Among cows with a CInt >13 months, mortality was higher in those with above-average yield at insemination compared to those with average or below-average yield.

5.2.2 Results from the high-producing herd subset

Among cows in the 150 most well-managed herds, defined as those with the highest average annual milk yield, the risk of on-farm mortality within 30 days after the third calving was slightly lower compared to the rest of the dataset (1.65% vs. 1.8%).

In this subset, no significant association was observed between CInt and on-farm mortality. There was also no significant effect found in the model including milk yield at the last insemination relative to the herd-average yield.

6. General discussion

6.1 Milk production

6.1.1 Milk production parameters

As expected, the extended VWP led to a longer CInt and lactation, which in turn resulted in a higher total MY during the second lactation compared with VWP50. In addition, the 305-day MY was higher in the VWP140 group, consistent with findings in both primiparous (Edvardsson Rasmussen et al., 2023) and multiparous cows (Niozas et al., 2019a). This difference in 305-day MY may partly be explained by the negative effect of pregnancy on daily MY (Hammond and Sanders, 1923; Strandberg and Lundberg, 1991), as cows in the VWP140 group likely experienced a less pronounced decline in yield owing to later conception.

The most notable finding was the similar average daily MY per day of CInt and per lactation day between groups. In our study, daily MY per day of CInt averaged approximately 34 kg ECM in both groups. This is consistent with findings from Arbel et al. (2001), who selected the highest-yielding half of cows within each herd, defined as those with a previous 305-day ECM above the herd average, and then randomly assigned them to either a conventional 60-day VWP or one extended to 120 days, and from Van Amburgh et al. (1997), who compared VWPs of 60 and 150 days, resulting in CInts of 13.2 and 16.5 months, respectively. Similarly, Österman and Bertilsson (2003) compared 12- and 18-month CInts in combination with two or three daily milkings in a research herd. They found no difference in daily MY per day of CInt, but cows in the 18-months treatment milked twice daily had a longer DPL compared with the other treatment groups. The similar daily MY observed across these studies, including ours, is likely explained by a higher proportion of lactating days and a lower proportion of dry days, which help sustain production per day of CInt despite longer CInt. In addition, delayed conception would be expected to postpone pregnancy-related decline in daily MY, thereby maintaining a high milk production in cows with extended VWPs. These dynamics likely act synergistically to produce similar daily MY across treatments.

In our study, daily MY per lactation day was not affected by VWP. In contrast, Arbel et al. (2001) reported that multiparous cows with a conventional 60-day VWP produced more ECM per lactation day than cows whose VWP was extended to 120 days (39.9 vs. 39.0 kg). A plausible explanation for this discrepancy includes differences in cow selection, as discussed above. In addition, the milk composition differed, with our cows having a higher milk solids content compared with the cows studied by Arbel et al. (2001). Because the proportion of milk solids tends to increase during the latter stages of lactation, this factor may have contributed more strongly to maintaining ECM yield in our study.

When comparing these studies, important differences in study design should be noted. In our study, all cows were included except those with a first-lactation 305-day yield below 70% of the herd average, representing about 1% of the data set. In contrast, Arbel et al. (2001) restricted participation to the highest-yielding half of cows within herds. Van Amburgh et al. (1997) primarily aimed to evaluate the effect of bovine somatotropin and the delayed influence of pregnancy on lactation persistency. Taken together, these aspects suggest that our study contributes particularly robust evidence in support of extended lactation as a viable management strategy.

In this thesis, milk yield was presented as 305-day yield as well as per day of CInt and per day of lactation in the second lactation. While all three measures were reported, the latter two provide a more robust basis for evaluating and comparing lactations of different lengths.

Milk yield near dry-off was slightly reduced in cows with extended CInts, from 33.7 to 32.5 kg ECM when the CInt was extended from 381 to 427 days, based on test-day yields 10 to 40 days before dry-off. This interval was selected to avoid including yields potentially influenced by management decisions immediately prior to dry-off. The relatively high MY at this stage, combined with the small difference between groups, indicates that cows managed with an extended VWP maintained good lactation persistency through to dry-off.

High MY at dry-off has been associated with negative effects on udder health, both during the dry period and in subsequent lactation (Bates and Dohoo, 2016). Given that the reduction in MY was relatively small, and occurred at a high production level, it is unlikely that the difference achieved in our study had any meaningful impact on udder health. Instead,

a smooth transition at dry-off relies on appropriate management interventions to suppress milk production in preparation for the dry period (Vilar and Rajala-Schultz, 2020).

When evaluating dry-off and DPL, certain settings can be applied in research herds but are challenging to implement in commercial herds. For example, in the study by Rehn et al. (2000), cows were dried off either six weeks before expected calving or when daily yield dropped below 5 kg ECM. Such thresholds are unlikely to be adopted in commercial herds due to practical and economic considerations, but the results provide valuable insights into lactation curve development. Rehn et al. (2000) reported shorter DPL in primiparous compared with multiparous cows, with no differences between treatment groups in the first lactation, but significantly longer DPLs in the VWP140 group than in the VWP50 group in later lactations. Research herds thus offer important opportunities to evaluate management strategies that may be difficult to test under commercial conditions.

In this study, DPL did not differ between treatment groups, and MY prior to dry-off was only marginally higher in the VWP50 group. These results suggest that a longer CInt did not increase the risk of an extended dry period due to early dry-off caused by low production in these commercial herds. Burgers et al. (2022) also reported no difference in DPL between treatment groups. In contrast, Lehmann et al. (2016) reported that extended lactations beyond a 13-month CInt increased DPL by 3-5 days.

Analyses of extended lactations based on retrospective records may yield biased results because cows with longer lactations are not randomly selected. According to Arbel et al. (2001), such studies are further limited by variability in milk yield across years, herds, climates, seasons and management practices.

6.2 Fertility

6.2.1 First service pregnancy and other fertility traits

In our study, there was a higher proportion of cows conceived at first service with an extended VWP compared with a conventional VWP, both among all inseminated cows and among those completing their second lactation. This result may be attributed to a longer recovery period after

calving, allowing cows to resume normal ovarian cyclicity before insemination. This interpretation is supported by Larsson and Berglund (2000), who, using progesterone profiles from the second week postpartum to monitor ovarian function, demonstrated that extending the VWP to 140 days reduced ovarian disturbances at insemination.

In contrast, a retrospective study of 11 Dutch commercial farms, where managers applied either individually tailored or fixed extended VWPs, reported variable and generally small effects on FSP (Burgers et al., 2021b). In that study, some managers extended VWP for all cows, others based their decisions on daily milk yield, delaying insemination until production dropped below a defined threshold, and others used peak yield as criterion. Such approaches may introduce bias because higher-yielding cows, which also tend to have genetically lower fertility, are more likely to have insemination postponed. As a result, these cows may appear to have poorer fertility outcomes, or no improvement, compared with cows in randomized trials where later insemination is not systematically linked to yield potential. These differences in study design and selection criteria likely contributed to the divergent findings in FSP between experimental and observational studies.

The higher FSP observed in our study contributed to fewer inseminations per conception (NINS) and a shorter interval from first to last insemination (IPL) compared with the conventional VWP group. These findings are consistent with results from research farms (Larsson and Berglund, 2000; Ma et al., 2022) and from commercial herds (Edvardsson Rasmussen et al., 2023). In Ma et al. (2022), the study's predefined insemination windows, up to 300 days in milk (**DIM**), resulted in insemination periods of 250, 175, and 100 days for VWPs of 50, 125, and 200 days, respectively. This design likely contributed to shorter IPL and lower NINS in the VWP 200 group. Conversely, Ratnayake et al. (1998) found no difference in NINS when comparing calving intervals of 12, 15, and 18 months, where cows were allowed up to five inseminations within 130 days post-first service.

Fixed limits on the number of inseminations or on the length of the insemination period are commonly applied in research herds, whereas in commercial herds they are less often used because of potential economic consequences. Research herd studies therefore provide valuable insights under controlled conditions, where fertility indicators such as NINS and

IPL can be clearly defined based on predetermined insemination limits or periods, and fertility-related culling can be consistently categorized. Commercial herd studies, in turn, capture the economic and practical realities of dairy farming. Together, these two research settings offer complementary evidence on the effects of extended VWP on fertility and culling.

First-service conception rate was identified as one of the best fertility indicators by Bertilsson et al. (1997). First service conception is normally defined as the proportion of cows confirmed to be pregnant after the first insemination, whereas in our study, FSP was defined more strictly as the proportion of cows that calved after only one insemination. A strength of these indicators is that they are measured early in the reproductive process and is not influenced by subsequent fertility events.

In commercial herd settings, decisions on whether to inseminate or not are rarely dictated by study protocols and are often influenced by managerial considerations, such as daily milk yield. Higher-yielding cows are generally more likely to be inseminated than lower-yielding cows. For high-yielding cows, DIM is also a less decisive factor in stopping insemination compared with low-yielding cows. These management practices may therefore contribute to differences in CFI, NINS, and IPL observed between VWP treatments (Eicker et al., 1996; Burgers et al., 2021b).

6.2.2 Voluntary waiting period and calving interval

In our study, extending the VWP did not result in a proportional increase of CInt; a 60-day extension of the VWP led to only a 46-day increase in CInt. This outcome indicates that improved fertility performance, particularly higher FSP, mitigated the expected lengthening of CInt. Notably, shorter VWPs may extend the interval between the end of VWP and CFI due to challenges such as anestrus and ovarian disorders in early lactation, as reported by Ratnayake et al. (1998). These findings indicate that VWP and CInt are not rigidly correlated, highlighting the importance of adjusting VWP according to herd-specific conditions when targeting a specific CInt. Furthermore, variability in estrus detection efficiency among farms can substantially influence the relationship between VWP and CInt, underscoring the need for tailored reproductive management strategies.

In our study, the intervals for CFI were 68 days in the VWP50 group and 127 days in the VWP140 group. The relatively low CFI in the extended group can partly be explained by cows being inseminated according to the study protocol. In addition, management traditions in Swedish dairy herds may have influenced the results; if a cow showed estrus less than half a cycle before the planned VWP, insemination was often carried out anyway. Thus, with a VWP set at 50 days, cows could occasionally already be inseminated from around day 40. In the VWP140 group, this practice was likely more frequent, as most cows had resumed cyclicity by then, meaning that up to half of the cows could potentially be inseminated from around 130 DIM, thereby contributing to the CFI ending up at 127 days in the VWP140 group.

As visualized in Figure 2, the VWP itself shaped the distribution of inseminations. In the VWP50 group, CFI was further prolonged by cows in anestrus, cows showing silent estrus, or cows for which insemination was deliberately delayed. As a result, the LSMean CFI was higher in the VWP50 group than would be expected based on the assigned waiting period alone. A similar pattern was observed in Arbel et al. (2001), where a VWP of 60 days resulted in a mean CFI of 71 days, while a VWP of 120 days yielded a mean CFI of 124 days. This likely reflects the fact that, by 120 days postpartum, nearly all cows had resumed estrus cyclicity, whereas at day 60, some cows in the VWP60 group were still in anestrus, thereby prolonging their CFI. In addition, cows that were not inseminated within 29 days after the assigned VWP were excluded from the analysis, which may also have influenced the results.

6.2.3 Culling

In our study, there was a tendency ($p = 0.07$) toward a higher risk of culling in the VWP140 group compared with VWP50. This difference could not be explained by cows culled without ever being inseminated or by cows reported as culled due to fertility problems. Similar patterns have been reported elsewhere: Niozas et al. (2019b) also observed a tendency toward higher culling rates in cows assigned to a VWP of 180 days compared with those assigned to 40 days. In contrast, Arbel et al. (2001) and Edvardsson Rasmussen et al. (2023) found no significant differences in culling rates between treatment groups. Interestingly, Larsson and Berglund (2000) reported more culling due to poor fertility, defined as failure to conceive

after five inseminations within 130 days, in the VWP50 group. It is not unlikely that culling patterns are influenced by the greater amount of information available at the time of first insemination when the VWP is extended. In our study, 1–2 additional test milkings provided further information about milk yield capacity and milk quality (e.g. SCC), which could affect decisions on whether to inseminate a cow. Extended VWPs may therefore enable more informed selection at the herd level. While conventional VWPs may result in pregnancies that in retrospect could be considered less desirable, extended VWPs create opportunities for more deliberate decisions, potentially leading to culling of cows that do not meet the farmer's expectations. Future studies should investigate whether such mechanisms contribute to differences in culling patterns between reproductive strategies.

6.3 On-farm mortality

6.3.1 Varying calving interval and on-farm mortality

The study focused on on-farm mortality within 30 days after the third calving. This approach was chosen to maintain homogeneity by studying the effect of an extended CInt during the second lactation. By restricting the analysis in this way, we also avoided potential bias that would arise in a retrospective study if cows of different parities were included, since mortality risk increases with advancing lactation number (Thomsen et al., 2004; Miller et al., 2008). Hypothetically, this risk could be further amplified by farmers' tendency to inseminate high-yielding or long-lived cows, even when their physical status may be declining, for example, cows approaching a lifetime production of 100 tons.

The intervals were chosen to reflect those commonly recommended in dairy consulting, corresponding to the control (11.7-13 months) and extended (13-15 months) CInt used in Paper 1, as well as shorter (<11.7 months) and longer (>15 months) alternatives. This categorization allowed us to compare mortality risk across a range of calving intervals that are both practically relevant and scientifically aligned with previous work.

The mortality within 30 days after third calving in our study was 1.78%. Thomsen et al. (2004) reported an average on-farm mortality risk of approximately 3.5% in 1999, with 30.5% deaths among cows in parities 1

and 2 and 41.1% among older cows occurring within 30 days of lactation. They also demonstrated an increased mortality risk with advancing parity. Thus, the level of early-lactation mortality observed in our study is comparable to that reported by Thomsen et al. (2004). Other studies also confirm the high risk associated with the onset of lactation; Miller et al. (2008) reported that 22.7% of deaths occurred within 45 DIM and Alvåsen et al. (2014b) found that 50% of deaths occurred within 78 days.

When modelling individual 305-day ECM yield, the probability of mortality within 30 days after calving increased with CInt longer than 13 months and was further elevated with CInt longer than 15 months compared with shorter intervals. Mortality risk also tended to increase with higher yield class. The intention with this model was to examine whether higher milk yield could provide some protection against mortality risk, for example by reducing the likelihood of over-conditioning, as fattening is a known risk factor for peripartum complications. However, the results did not confirm this hypothesis. Moreover, because 305-day yield is standardized to a 12-month CInt and tends to increase with longer intervals, it may be a misleading measure in this context.

Instead, we modelled daily MY at the time of insemination relative to the herd-average MY at the third test milking within a year. Using this strategy, cows were categorized into three groups: within ± 2 kg ECM of the herd average, above this threshold, and below it. The aim was to create groups with differing risks of body condition gain from conception to dry-off. The results showed that despite adjusting for MY at insemination, longer CInt was still associated with an increased risk of mortality. In addition, cows with higher yields at conception and a longer CInt had a higher mortality risk compared with those with lower yield, which is consistent with the findings of Pinedo and De Vries (2010). They reported a doubled risk of mortality in cows with extended days open (90–300 days) compared with shorter intervals and also found a higher mortality risk in cows with lower yields at dry-off.

Calving interval may serve as a proxy for overall herd management, where farmers achieving short CInts may also excel in practices such as maintaining good cow condition and exercising sound judgment about when to refrain from inseminating a cow and instead cull her. In our study, we further developed the model by focusing on the 150 highest-yielding herds, under the assumption that high-producing herds generally represent

better management than lower-producing herds. Herds were ranked by average milk yield, and the aim was to reduce the proxy effect, ensuring that cows with extended CInt were more likely to represent deliberate management choices rather than suboptimal management. In this subset, a numerically, but not statistically significant, increase in mortality risk with longer CInt was observed among cows producing more than 2 kg ECM less than the herd average at the last insemination. For cows in the average and high-yield groups, no clear trend was detected, and the difference in mortality risk between the shortest and longest CInt classes was smaller compared with the full dataset. Overall mortality was also lower in these well-managed, high-yielding herds (1.65%) than in the remaining herds (1.80%). These findings support the hypothesis of Alvåsen et al. (2014a) and Shahid et al. (2015) that the increased risk of mortality in herds with long CInts is associated with suboptimal management rather than a direct causal effect. It is also important to acknowledge a key limitation of this retrospective study, we cannot determine whether extended CInt resulted from voluntary management decisions or from involuntary factors such as fertility problems or health issues, as previously discussed. This uncertainty highlights the need for cautious interpretation and for further studies.

As mentioned, several studies have reported an increasing trend in on-farm mortality and have identified a number of associated risk factors. To counteract this negative development, more knowledge about the underlying causal relationships is needed. Our results indicate an increased risk of on-farm mortality from a CInt of 13 months, and particularly beyond 15 months, while more skilled farmers, represented here by herds with higher average MY, appear to face a less pronounced risk compared with the average herd. Control schemes are required to reduce economic and animal welfare costs as well as to manage reputational risks posed to the dairy industry (Compton et al., 2017). One possible draft for such a scheme, based on our results, would be to require a critical assessment of cow condition before inseminating cows in estrus later than 180 DIM, especially if higher on-farm mortality risk can be anticipated. According to Compton et al. (2017), no reports of successfully implemented control programs targeting dairy cow mortality have been published.

6.4 General considerations and reflections

6.4.1 Fertility strategy

By including today's high-yielding herds, this study provides evidence-based support for best practice in modern dairy production. Given the ongoing trend of increasing milk yield, these herds are also likely to represent tomorrow's average-producing herds, making the findings relevant for both current and future management strategies, as well as a foundation for decision support.

As noted previously, average annual MY varies considerably between herds. High-performing herds often benefit from superior nutrition, cow comfort, and overall management, which not only support higher peak yields but may also enhance lactation persistency. Under such conditions, extended VWPs are likely to be more feasible and beneficial for multiparous cows. As discussed, these well-managed herds also appear to be able to implement extended CInt without an increased risk of on-farm mortality at subsequent calving, in contrast to average and low-yielding herds. In many of these high-performing herds, the average 305-day yield already exceed 14,000 kg ECM, and individual cows may surpass 16,000 kg ECM. As the number of such high-yielding cows continues to increase, there is a growing need to understand their lactation and reproductive dynamics. Such knowledge will be essential for developing more refined, herd-specific reproductive strategies, moving beyond current one-size-fits-all VWP recommendations.

Overton et al. (2025) argued that, given the focus of modern dairy farming on maximizing genetic potential and productivity, striving for reduced production seems illogical. However, results from our study, as well as other studies discussed above, demonstrate that extended lactations can maintain production while improving fertility outcomes and possibly offering advantages at dry-off. Moreover, ongoing genetic selection for higher yield also favors lactation persistency, suggesting that the potential benefits of extended voluntary waiting periods may increase in the future. The development of decision-support tools; drawing on milk yield data, genomic information, and increasingly advanced AI systems integrated with national databases such as SOMRS and herd management software, could further support optimization of VWP, reducing the burden on farmers to judge each cow's optimal timing individually.

6.4.2 Compliance with the protocol

To address protocol deviations, analyses can be conducted either per protocol, which would include only compliant animals but risk bias if exclusions are systematic, or intention to treat, which includes all randomized animals and thereby better reflects real-world outcomes, though it requires adequate compliance (Tripepi et al., 2020). In our study, the aim was to include all cows in their assigned treatment group. The only exception was the exclusion of cows with a first-lactation 305-day MY below 70% of the herd average. This rule was implemented both to build trust among farmers by signalling that the trial focused on high-potential animals and to minimize the risk that inclusion of very low-yielding cows would result in management challenges, such as over-conditioned animals or extended dry periods. It also reduced the incentive for farmers to exclude low-potential cows on their own initiative. Despite these measures, three herds were excluded due to poor compliance: two herds displayed a seasonal pattern of adhering or not adhering to the protocol, and one herd tended to systematically exclude Swedish red cows from the extended group.

7. Conclusions

The studies included in this Licentiate work provide the following main conclusions:

- In second-parity cows, an extended voluntary waiting period did not affect daily milk yield per calving interval day, milk yield per lactating day or dry-period length.
- Fertility improved with extended voluntary waiting period, as indicated by a higher first service conception.
- No effect of extended voluntary waiting period on culling rate was detected.
- Mortality risk was associated with high milk yield, measured both as 305-day yield and as yield at the last insemination relative to the herd average at the third test milking. In the subset of the highest-yielding herds, these associations were not statistically significant.

8. Future research

Future research is desirable, since extended lactations present certain advantages but also carry potential risks that must be carefully considered.

- Further development of models evaluating varying voluntary waiting periods at the cow and herd level, including effects on milk yield, lifetime production, and milk yield per cow per day at the herd level. Such models should also incorporate overall impacts on the proportion of lactating and dry cows, and the use of feed resources, barn space and labor as well as environmental effects.
- Further development of models assessing disease frequency, culling rate, and on-farm mortality, with calving intervals adjusted to better reflect practical herd management and current knowledge needs.
- Continued research on mortality, with the aim to identify the underlying causes of increased mortality risk observed with extended lactation.

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Popular science summary

Traditionally, Swedish dairy producers have aimed for a calving interval of 12–13 months, meaning that each cow calves once per year. As milk yields has increased over time, interest in extended calving intervals has grown. For many farmers, drying off high-yielding cows is a challenge, and statistics show that the highest risk of disease and mortality occurs around calving. A longer interval between calvings, may potentially reduce the number of health problems for the cow.

Earlier studies have shown that first-parity cows are particularly suited for longer lactations, since they typically have a more even milk production level over time. In this study, we instead focused on second-parity cows, which have an overall higher milk yield but a more rapid decline of the milk production in the later stages of lactation. The study was carried out in 12 high-yielding herds with different production conditions, including feeding strategies, conventional or automatic milking systems, and two or three milkings per day.

Cows were either inseminated about 50 or 140 days after calving. The results showed that cows with a longer waiting period before insemination, and thereby longer calving interval, had better fertility outcomes. The results suggest that cows with a longer waiting period, and thus recovery period, before insemination can be beneficial. Milk production between two calvings was not affected by the length of the calving interval.

Data from the Swedish official milk recording system over a seven-year period were analysed in a second, retrospective study. These results showed that the risk of mortality after calving increased for cows with longer calving intervals. However, this association was not found in the subset of the highest-yielding herds. The results of such retrospective studies should be interpreted with caution, since a cow may have received a longer calving interval for other reasons, such as illness or reduced fertility, which in turn could have affected the mortality risk.

In summary, the findings suggest that longer calving intervals can be a valuable alternative to the traditional 12-month cycle, particularly in high-yielding herds. At the same time, our analyses of historical data show an association between longer calving intervals and a higher risk of mortality around calving.

Populärvetenskaplig sammanfattning

Traditionellt har svenska mjölkproducenter siktat på att korna ska kalva en gång per år, vilket givit ett kalvningsintervall på 12–13 månader. Men med en ökad mjölkavkastning har intresset för längre kalvningsintervall ökat. För många lantbrukare är det en utmaning att sinlägga högproducerande kor, och statistik visar att störst risk för sjukdom och dödsfall är i samband med kalvning. Med färre kalvningar finns det en potential att minska frekvensen av hälsoproblem för mjölkkorna.

Tidigare studier har visat att förstakalvare ofta passar bra för längre laktationer eftersom de har en mer jämn mjölkproduktion över tid. Denna studie har i stället fokuserat på andrakalvare, som har en högre mjölkproduktionskapacitet, men har en snabbare minskning av mjölmängden senare delen av laktationen. Den första studien genomfördes i 12 högavkastande besättningar med olika produktionsförutsättningar som t.ex. olika utfodringsstrategier, traditionell eller robotmjölkning och med mjölkning två eller tre gånger per dag.

I studien sminerades korna vid olika tidpunkter efter kalvning, antingen efter 50 dagar eller efter 140 dagar. Resultaten visade att en senarelagd första sminering efter kalvning gav bättre fruktsamhetsresultat, vilket kan tolkas som att en längre återhämningsperiod efter kalvning är positivt. Mjölkavkastningen påverkades inte av kalvningsintervallens längd.

Data från den svenska officiella kokontrollen under en sjuårsperiod analyserades i en andra studie. Där framkom det att risken för dödlighet i samband med kalvning ökade för kor med längre kalvningsintervall. I ett urval av de mest högavkastande besättningarna kunde detta samband inte ses. Samband baserade på historiska data måste tolkas med försiktighet. Kon kan ha fått ett längre kalvningsintervall av olika skäl, till exempel på grund av sjukdom eller nedsatt fertilitet, som i sin tur kan påverka riskerna vid kalvning.

Sammanfattningsvis visar resultaten att längre kalvningsintervall kan vara ett fullgott alternativ till det traditionella 12-månadersintervallet, särskilt i högavkastande besättningar. Samtidigt visar våra studier på historiska data att det finns ett samband mellan längre kalvningsintervall och en ökad dödlighet kring kalvning.

Acknowledgements

This project originated over a dinner conversation at a conference in Umeå, where I happened to be seated next to Professor Kjell Holtenius. We discussed optimal calving intervals, and I shared my thoughts and experiences with extended voluntary waiting periods in older cows in high-yielding herds from my work as a dairy consultant at Växa. He encouraged the idea and subsequently prepared and submitted an application to the Swedish Foundation for Strategic Research, which awarded a grant (SM18-0042) that, together with support from Växa Sverige and the Swedish Dairy Association enabled these studies as an industrial licentiate project. Additional travel grants from Olof Claessons Fond and the Royal Swedish Academy of Agriculture and Forestry (KSLA) made it possible for me to attend and present preliminary results at EAAP conferences in Porto and Lyon.

The journey from initial ideas to scientific knowledge would not have been possible without my outstanding group of supervisors. I am deeply grateful to Kjell (Leo) Holtenius, Mikaela Lindberg, Renée Båge, and Cecilia Kronqvist for their guidance, encouragement, and support.

Leo, my main supervisor for the first article, thank you for your scientific and project guidance. I had the privilege of working with you during the most experience-rich phase of your distinguished career. I will now be your latest student—and likely the last—you have supervised. The many discussions at Ultuna, often during fika or breaks, revealed the enormous breadth of your knowledge. Sometimes I was actively involved, and other times I simply listened, but in every case I learned from the insights you so generously shared with us.

Mikaela, my main supervisor for the second article, it has been an absolute privilege to be guided and supported by you. Your ability to balance the topics and elevate the texts has greatly improved the final results. I have also benefited from your exemplary leadership, and from the countless variations of “Kämpa på!” you have sent me. In my eyes, you are a true role model, and beyond the scientific knowledge, your example is something I will carry with me and strive to emulate in my future.

Renée, thank you for evaluating and assessing the reproductive findings in this study. You brought a unique ability to combine deep reproductive

research expertise with practical outcomes and applications. You argued for the inclusion of variables that added important perspectives, and without the time limits of this project, your many ideas for further studies could easily have kept it going much longer. Above all, your genuine passion for science has been inspiring throughout this journey.

Cicci, what a privilege it has been to benefit not only from your expertise in statistics and your skills in SAS, which proved invaluable when working with data from the cow database, but also from your broad scientific knowledge. It has been truly enriching to receive your insightful—and often witty—comments on my manuscripts. They challenged me to reconsider aspects I thought were already clear, which made the work both more rigorous and more rewarding.

Anna, my always caring, warm, supportive, and truly generous companion in entering the world of scientific work. I have been so lucky that you were ahead of me, allowing me the advantage of following your path. You have never left a friend in need without help, and I am deeply grateful for that.

This work was carried out at the Department of Applied Animal Science and Welfare, SLU, and at Växa. It was formally signed by Sara Österman, Head of Department, and Sara Lundberg, Växa Sverige. The project was further supported and formally endorsed by Fredrik von Unge, Head of Knowledge, Research and Development at Växa.

Under studietiden fick jag möjlighet att prova på biträdande handledarskap. Tack för förtroendet, Jan-Eric och Cicci. Tack också till studenterna, Viktor Ronström och Malin Areskoug, som valde att skriva sitt examensarbete om det intressanta området kors kalvningsintervall. Och till Arvid Idhamre och Nathalie Persson, som skrev sitt examensarbete om förlängt kalvningsintervall, där jag hade förmånen att vara en av de intervjuade.

På SLU har jag haft förmånen att få guidning i statistikens fantastiska värld av Jan-Erik E. och Adam F. Tack också till Sigrid A. för kloka ord och uppmuntran, till Rebecca D. för att du delat med dig av dina erfarenheter som doktorand, och till Ewa W. för din värme och omtanke.

Tack Karin A. för din tid, ditt lugn och ditt tålamod i våra diskussioner kring kodödlighet. Ett tack även till Tomas K. och Mårten H. för er smittande energi och ert aldrig sinande flöde av idéer om framtida möjligheter. Till sist, ett tack till Johan K. och Sonia S. för er hjälpsamhet med boende.

Från Växa har Sara Lundberg stöttat arbetet från allra första början och deltagit i referensgruppsmöten för att följa projektets utveckling. Mina två närmaste chefer, nuvarande Malin Stiernstedt och tidigare Helen Börjesson, har följt med på resan. Ett varmt tack för ert stöd och er uppmuntran! Tack även till Anki Roth för dina reflekterande inlägg och insiktsfulla diskussioner.

Tack också Växa Sverige och de kokontrollanslutna gårdarna för datamaterialet. Ett särskilt tack riktas till Nils-Erik Larsson och Staffan Ericsson för era värdefulla insatser vid datahanteringen.

Stort tack till alla djurägare och all personal som antog utmaningen att delta som försöksgårdar. Ert mod och engagemang, tillsammans med alla givande diskussioner och lärdomar, har varit ovärderliga.

The THV PhD fellows—always ready with smiles, chats, board games, pep talks, and pizzas. Thank you for sharing your experiences and for building a network that spans the world. It has truly been a pleasure to meet you all.

Ett stort tack till nuvarande och tidigare kollegor. Ett särskilt tack till Susanne för hjälp med ansökan och rekvirering, och till Sara Bergström för alla fyndiga och peppande hundpromenader – du har hjälpt mig att hålla lågan och hoppet vid liv. Tack Maria L. för ditt osvikliga stöd och dina professionella insatser. Katarina och Tellus – många rundor har det blivit med samtal både om R och om när man ska seminera. (Blir det bara prat om krämpor när detta är klart?) Helena L. introducerade mig till ChatGPTs underbara värld – där har vi numera en gemensam vän. Tack Emelie O. för alla dina lösningar, för din energi och för att du återintroducerade ridningen i mitt liv. Och tack bästa Ann N., alltid vänlig, trygg och stabil, ända sedan

agronomtiden på Ultuna. Till sist, Jan S., Ann-Christin S. och Kristina R. – dyker det upp frågor om kokontrollen så vet man att ni löser det.

Till alla engagerade tekniker, fruktsamhetsrådgivare, sektionsförmän, avelsrådgivare och veterinärer jag stött på före, under och efter fältstudien. Er nyfikenhet, ert ifrågasättande och era kloka kommentarer har berikat mig och gjort detta arbete bättre. Ett varmt tack till er alla - ingen nämnd ingen glömd!

Och tack till mina goda vänner i Uppsala, Bosse och Maggan, som har följt mig sedan tiden som student på agronomprogrammet. Maggan gick tyvärr bort innan denna avhandling blev klar, men min tacksamhet för er båda och för ert alltid öppna hem, våra långa samtal och goda måltider består.

Torbjörn Elvius, som tyvärr gick bort under arbetets gång. Tack för vänskap och för alla pauser vid fika, middagar och utflykter.

Sist men inte minst, tack till min älskade och stöttande familj!

Mamma – för att du alltid finns där, för din uppbackning, ditt öppna hem, våra dagliga samtal om stort och smått, och för att du alltid ställer upp i alla lägen. Jag kan inte tacka dig nog.

Magnus, Helena och Axel – för alla samtal, träffar och inte minst slutsatsen: *”ökar det så ökar det, minskar det så minskar det.”*

Anders och Simon – Simon, du gick i högstadiet när studien började, och nu har du fått jobb och flyttat hemifrån. Anders, du har hållit ut, och nu är det snart klart. Tack för allt!

This thesis evaluated extended voluntary waiting periods in high-yielding dairy cows. In a randomized controlled trial, a 140-day VWP improved fertility without reducing milk yield or affecting culling. A retrospective cohort study showed that calving intervals exceeding 13 months were associated with increased mortality risk, especially in high-yielding cows. Overall, extended VWPs may enhance fertility, but longer intervals can increase mortality risk, highlighting the need to balance reproductive strategies in modern dairy herds.

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ISBN (print version) 978-91-8124-023-8

ISBN (electronic version) 978-91-8124-024-5