CHALLENGING THE PARADIGM OF ONE CALF PER YEAR

Consequences of extended lactations for individual dairy cows

Eline Burgers

Propositions

1. Extending the lactation length is beneficial for primiparous cows and farmers.
(this thesis)
2. All multiparous cows need a customized lactation length.
(this thesis)
3. Denying anthropomorphism is as objectionable as promoting it is.
4. Supermarkets exploit customers who value animal welfare.
5. You do not have to comprehend music to benefit from it.
6. Getting lost on purpose leads to unexpected discoveries.
7. A calf a year keeps the vet near.

Propositions belonging to the thesis, entitled

Challenging the paradigm of one calf per year. Consequences of extended lactations for individual dairy cows.

Eline Burgers
Wageningen, 30 September 2022

Challenging the paradigm of one calf per year

Consequences of extended lactations for individual dairy cows

Eline Burgers

Thesis Committee

Promotors
Prof. Dr B. Kemp
Professor of Adaptation Physiology
Wageningen University & Research

Prof. Dr H. Hogeveen
Personal chair at the Business Economics Group
Wageningen University & Research

Co-promotor
Dr A.T.M. van Knegsel
Senior Researcher, Adaptation Physiology Group
Wageningen University & Research

Other members
Prof. Dr R.F. Veerkamp, Wageningen University & Research
Prof. Dr U. Emanuelson, Swedish University of Agricultural Sciences, Uppsala, Sweden
Dr A. Römer, Institute for Animal Husbandry, State Research Centre for Agriculture and Fisheries, Mecklenburg-Vorpommern, Germany
Dr W. Steeneveld, Utrecht University

This research was conducted under the auspices of the Graduate School Wageningen Institute of Animal Sciences.

Challenging the paradigm of one calf per year

Consequences of extended lactations for individual dairy cows

Eline Burgers

Thesis
submitted in fulfilment of the requirements for the degree of doctor
at Wageningen University,
by the authority of the Rector Magnificus,
Prof. Dr A.P.J. Mol,
in the presence of the
Thesis Committee appointed by the Academic Board
to be defended in public
on Friday 30 September 2022
at 1:30 p.m. in the Omnia Auditorium.

Eline Burgers

Challenging the paradigm of one calf per year. Consequences of extended lactations for individual dairy cows. 212 pages.

PhD thesis, Wageningen University, Wageningen, the Netherlands (2022)
With references, with summaries in English and in Dutch

ISBN: 978-94-6447-314-8
DOI: https://doi.org/10.18174/574043

Abstract

In most dairy systems, cows are managed to have a calf every year. A yearly calving is expected to maximize milk production due to the related yearly lactation peak. During the transitions around calving, cows have an increased risk for diseases. Lactations could be deliberately extended by extending the voluntary waiting period for insemination (VWP). This increases the calving interval (CInt) and reduces the frequency of calvings. A longer lactation and longer period in late lactation, however, may be related with a lower milk production, and a risk for fattening at the end of the lactation. This project aimed to evaluate the consequences of extended lactations for milk production, fertility, health, metabolism, and economic performance of individual dairy cows, by using both data from commercial farms and an experimental approach. Additionally, the aim was to identify cow factors that determine the response of individual cows to an extended lactation. At 13 commercial dairy farms where the VWP was deliberately extended for part of the herd, farmers selected their high-producing cows for an extended lactation, as indicated by the greatest 305-d production in the groups with the longest CInt. Milk production per day of CInt, however, was not always greatest for these cows. A longer CInt, but not a longer calving to first service interval, was related with an increased number of inseminations. In an experimental setting, the VWP was extended until 125 or 200 days for 41 primiparous (PP) and 113 multiparous (MP) cows. The VWP could be extended until 200 days for PP cows and until 125 days for MP cows with no effect for milk production per day of CInt. Multiparous cows with a VWP of 200 days had a lower milk production per day of CInt compared with MP cows with a shorter VWP. The lactation persistency between day 100 in lactation and the start of dry-off was improved for cows with a VWP of 200 days compared with cows with a VWP of 50 days. At the end of the extended lactation, an extended VWP resulted in a lower milk production at dry-off, which maybe be beneficial for udder health, but was also related with body fattening of MP cows. At the start of the next lactation, MP cows with a VWP of 200 days had a greater body condition score, a more severe negative energy balance, and a greater plasma non-esterified fatty acid concentration compared with MP cows with a VWP of 50 days, indicating an increased risk for metabolic disorders in the next lactation. For PP cows, the VWP did not affect milk yield or body condition at the end of the lactation or metabolic status at the start of the next lactation. Therefore, PP cows and MP cows with a high milk production and a low body condition before insemination may be more suitable for an extended VWP. Cows with a VWP of 200 days had lower total yearly revenues, but also lower total yearly costs, compared with cows with a VWP of 50 days. The yearly net partial cashflow was not significantly associated with the VWP, possibly partly due to the great variation among cows. To conclude, primiparous cows can handle an extended lactation very well, as an extended VWP did not affect their milk production or metabolism. For multiparous cows, a customized lactation length may limit the risk for a low milk production and fattening at the end of the extended lactation.

Contents

Chapter 1

General introduction

1 Traditional lactation length of dairy cows

In most dairy systems, cows are usually managed to have a calf every year. Evolutionary, a one-year calving interval (CInt) makes sense as it ensues from the seasonal calving in nature. In nature, calves are born in spring, when nutrients are abundant in the environment. In pastoral dairy systems, where cows are still highly dependent on nutrient availability from the environment, a seasonal calving is usually implemented (Dillon et al., 1995; Butler et al., 2010). In most modern dairy systems, year-round availability of silages and concentrates lowers the dependence on fresh nutrition from the environment. In addition, fresh milk production year-round may be beneficial and seasonal additional allowances used to exist to support this. As such, in most intensive or semi-pastoral dairy systems calves are born throughout the year. Most dairy farmers, however, still aim for a short CInt of about one year. The reason behind this is that a yearly calving is expected to maximize yearly milk production due to the associated yearly peak in milk production in the beginning of every new lactation (Dijkhuizen et al., 1985; Steeneveld and Hogeveen, 2012).

1.1 Issues with a traditional lactation length

Considering cow health, calving is a challenging process, due to the process of parturition and the start of a new lactation. The stress of parturition imposes a physiological challenge for a dairy cow and is related to an increased susceptibility to diseases (Goff and Horst, 1997; Kimura et al., 2006). Moreover, the start of a new lactation is characterized by a high peak in milk production (Butler et al., 1981; Rastani et al., 2005). Selection for a greater milk production has been related with increased feed intake, but not enough to sustain the milk production (Veerkamp et al., 2003). The steep increase in milk production towards the high peak milk production, together with a limited nutrient intake during this time, usually results in a negative energy balance (NEB) in the first 8-10 weeks of lactation (de Vries et al., 1999; Butler, 2005). This NEB is related with metabolic disorders, such as hypocalcemia and ketosis (Ingvartsen et al., 2003; Friggens et al., 2004). An NEB and ketosis have been associated with impaired immune function related with increased susceptibility to infections in early lactation. In addition, the start of a new lactation is specifically related with a reduced mammary protection against exogenous bacteria, due to the loss of the keratin plug that seals the teat (Goff and Horst, 1997). Moreover, in case of dry cow therapy, at the start of the lactation the protective function of these dry cow antibiotics disappears, which could increase the risk for mastitis. Due to the parturition process and the increased risk for diseases related with the start of a new lactation, most culling as a result of disease happens in the first 8 weeks of lactation (Fetrow et al., 2006; Dechow and Goodling, 2008; Pinedo et al., 2014). Events during the first 8 weeks of lactation therefore substantially contribute to the lifespan of dairy cows.

Concerning consequences of a traditional lactation length for calves born from dairy cows, in the Netherlands, on average 30% of these calves are kept for replacement (CRV, 2020), and around 70% are raised for meat consumption, meaning they usually leave the dairy farm at 14 days of age. These calves are perceived as a by-product of the dairy industry and have a limited value for the dairy farmer (Mohd Nor et al., 2012). Moreover, the public is concerned with potential issues regarding calf welfare and health in the veal industry, which is an important market for surplus calves born on dairy farms (Bokma et al., 2020). Considering consequences of a one-year CInt for cow health, lifespan, and surplus calves, it can be questioned if these yearly calvings are necessary and desirable for all dairy cows.

2 Extended lactation length

Lactations could be deliberately extended by extending the voluntary waiting period for insemination (VWP). This increases the calving interval and reduces the frequency of calvings (Figure 1). A lower frequency of calvings for cows results in a lower frequency of critical transitions related with calving, possibly improving cow health and reducing the risk for culling due to disease, which could increase the cow's lifespan. In addition, with a lower frequency of calvings, the yearly number of surplus calves is reduced. Finally, an extended lactation may improve production efficiency, as the number of days dry relative to the lactating period and the input of concentrates relative to the total milk produced might be reduced (Knight, 2005; Lehmann et al., 2014). Moreover, if extended lactations reduce the disease incidence, fewer days of disease treatments may reduce discarded milk and herewith improve the production efficiency. Extended lactations thus have the potential to contribute to improved animal health, increased lifespan, reduced number of surplus calves, and increased production efficiency.

Extended lactations, however, could also have disadvantages. When the frequency of calving moments is reduced, cows have a lower frequency of peaks in milk production, and spend relatively more time in late lactation, where milk production usually is lower. When extended lactations are applied on farms, this can be hypothesized to lead to a reduction in milk on a herd level. A lower milk production could reduce the farmer's income and increase the environmental impact per kg milk (Lehmann et al., 2019; Kok et al., 2019). Moreover, as in late lactation milk production usually is reduced, cows can be expected to have an increased risk for fattening at the end of the extended lactation. In the next sections, consequences of extended lactations for milk production and persistency, fertility, metabolism and health, and farmer's income are discussed, as well as their relationships with individual cow characteristics.

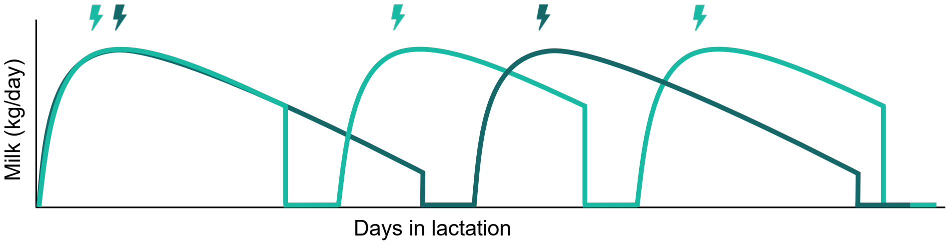


Figure 1. Three lactation curves in a one-year calving interval (CInt; light lines) and two lactation curves in an extended CInt (dark lines), assuming no effect of pregnancy on the lactation curve. For the one-year CInt, the dry period starts at a higher production level, the frequency of peaks in milk production is greater, the frequency of transitions around calving (⚡) is greater, and the dry period relative to the lactation is longer compared with the extended CInt.

2.1 Consequences for milk production and persistency

Cows with an extended lactation have a lower frequency of peaks in milk production, and spend more time in late lactation, when milk production usually is lower (Strandberg and Oltenacu, 1989). When the length of the dry period remains the same, however, an extended lactation results in fewer days dry relative to days in lactation. Most studies that modeled the effect of extended lactations on milk production reported a reduction in milk production in longer CInt (Steeneveld and Hogeveen, 2012; Kok et al., 2019). These modeling studies, however, used retrospective farm data or effect studies as input, where lactations mostly were not deliberately extended as a strategy of the farmer. A recent observational study investigated retrospective data of commercial dairy farms that deliberately extended the lactation of their cows (Lehmann et al., 2016). To compare milk production in the different CInt, milk production per day of CInt was calculated as the milk production in the complete lactation divided by the days of the CInt, including the dry period. In that study, milk production per day of CInt increased when the CInt was longer, possibly related with a selection of high-producing cows for longer lactations by the farmers and proportionally fewer days dry relative to days in milk per CInt (Lehmann et al., 2016). In controlled experiments, the effect of a deliberately extended lactation on milk production varied among studies. Some studies reported no significant effect of the VWP on milk production per day of lactation (Österman and Bertilsson, 2003; Niozas et al., 2019a) or a tendency for a lower milk production per day of CInt when the VWP was extended (Rehn et al., 2000). Differences in extension of the VWP or differences in cow characteristics among studies could have affected the consequences for the milk production. For example, in the study by Rehn et al. (2000), numerically the milk production per day of CInt increased for primiparous cows and decreased for multiparous cows when the VWP was extended from 50 to 140 days. Moreover, another study only enrolled high-producing cows, defined as a primiparous cow with once a milk production of at least 30 kg per day in the first 3 monthly records or a multiparous cow with an above herd average 305-d production in the previous lactation. In that study, primiparous cows had a greater value corrected milk production per day of CInt when the VWP was extended from 90 to 150 days, and for multiparous cows the VWP did not affect the milk production when the VWP was extended from 60 to 120 days (Arbel et al., 2001). Next to variation among cows, also management strategies may affect the consequence of an extended lactation for milk production. For example, in the study of Österman and Bertilsson (2003), cows with a VWP of 50 or 230 days had the same milk production when they were all milked 3 times per day (Österman and Bertilsson, 2003). Moreover, with a VWP of 230 days, milk production increased when cows in this group were milked 3 times per day compared with milking 2 times per day (Österman and Bertilsson, 2003).

Persistent lactations can reduce the risk for a low milk production at the end of the lactation. Several studies reported a negative effect of pregnancy on persistency (Erb et al., 1952; Olori et al., 1997; Brotherstone et al., 2004; Yart et al., 2013). As in an extended lactation the gestation is delayed, persistency may be expected to be improved. Indeed, lactation persistency as the rate of decline in milk production per day was increased from -0.071 to -0.063 or -0.061 kg/day with an extension of the VWP from 40 to 120 or 180 days in milk (DIM) (Niozas et al., 2019a). In addition, parity might affect persistency of cows in an extended lactation, as usually primiparous cows have more persistent lactation curves compared with multiparous cows (Arbel et al., 2001; Lehmann et al., 2016; Niozas et al., 2019a).

2.2 Consequences for fertility

When cows are managed for a one-year CInt, they need to become pregnant around 85 DIM. In practice, the VWP is usually 40 to 60 days, implying a CInt between 320 and 350 days if the first insemination is successful. In contrast, the average calving interval in Dutch dairy farms was 407 days in 2020 (Figure 2; CRV, 2021), implying that cows conceive at on average 127 DIM.

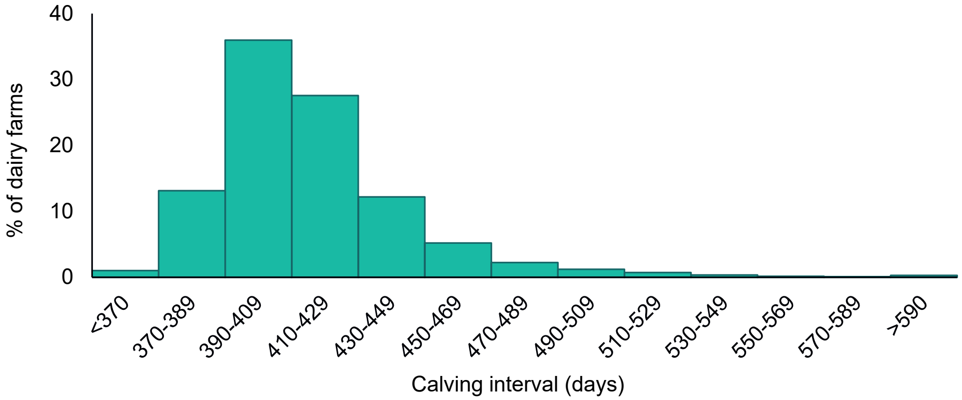


Figure 2. Percentage of dairy farms per average calving interval in the period between September 2019 and August 2020 (adapted from CRV, 2021).

A possible explanation for the impaired reproductive performance in early lactation is that cows are still in an NEB during that period (Opsomer et al., 2000; Leroy et al., 2008). In nature, a severe NEB as a result of lactation and poor nutritional conditions may be related with reduced pregnancy rates (Knight, 2001; Hackmann and Spain, 2010). The underlying trade-off between investment in the viability of the current calf and investment in future offspring may imply that cows with a high milk production and a low feed intake resulting in an NEB have a reduced reproductive performance at that moment, as the considerable investment for a new calf makes reproduction a high risk in harsh nutritional conditions (Friggens et al., 2010).

In several studies, the relation between the NEB and reproductive performance has been investigated (Opsomer et al., 2000; Jorritsma et al., 2004; Leroy et al., 2008). The NEB is associated with an altered metabolic status reflected by a decreased plasma glucose, insulin, and insulin-like growth factor 1 (IGF-1) concentration and an increased plasma non-esterified fatty acid (NEFA) and β-hydroxybutyrate (BHB) concentration, which have been related with an impaired maturation and development of oocytes (Jorritsma et al., 2004; Leroy et al., 2006) and an altered uterine environment which may impair embryonic development (Wathes et al., 2003). In dairy systems, next to the milk production level and related NEB of the cows, also management has an influence on reproductive successes of cows (Walsh et al., 2011; Rethmeier et al., 2019). For example, herds with high productions (>35 kg milk per cow per day) had a similar reproductive performance as herds with low productions (<30 kg milk per cow per day). This may indicate that not milk production per se but also management affects reproductive performance (Rethmeier et al., 2019). In that study, however, the management in the different herds also may have affected the milk production. Moreover, within these herds, possibly there still may have been a difference in reproductive performance between lower and higher producing cows. Overall, on a cow level, a high milk production and NEB are related with reduced reproductive performance (Lucy, 2001; Butler, 2005; Chen et al., 2015).

When the lactation is extended by deliberately delaying the insemination, start of insemination takes place later in lactation. Later in lactation, milk production can be expected to be decreased (Gaillard et al., 2016), and feed intake to be increased. As a result, the energy balance (EB) is improved (de Vries et al., 1999). A more positive EB at the moment of insemination could improve reproductive performance (Leroy et al., 2008). This could reduce costs for multiple inseminations that may be necessary when starting early in lactation to impregnate a cow, and moreover could reduce culling due to fertility problems when inseminating later in lactation. In an observational study at commercial farms where farmers deliberately extended the CInt, CInt length was not related with conception rate (Lehmann et al., 2016). This may be explained by the selection of high-producing cows for a delayed insemination at those farms, possibly resulting in a similar EB and reproductive performance at the time of insemination. The effect of a delayed insemination on reproductive performance when cows were randomly assigned to different VWP varied among studies (Schneider et al., 1981; Schindler et al., 1991; Bertilsson et al., 1997; Van Amburgh et al., 1997; Arbel et al., 2001; Niozas et al., 2019b). One study reported an increase in services to conception from 1.50 to 1.95 when the insemination was delayed from 50 to 80 days (Schneider et al., 1981). More recent studies did not report a reduced reproductive success when the insemination was deliberately delayed. Some studies reported no effect of an extended VWP on reproductive performance when the VWP was extended from 60 to 150 days (Van Amburgh et al., 1997) or when cows were managed for a CInt of 12 or 15 months (Bertilsson et al., 1997). Other studies reported a better reproductive performance when the VWP was extended. For example, one study reported fewer services to conception and fewer days open when the VWP was extended from 40 to 120 or 180 days (Niozas et al., 2019b).

Moreover, multiparous cows, but not primiparous cows, had a greater conception rate at first insemination when cows were inseminated between 120 and 150 DIM instead of before 90 DIM (Schindler et al., 1991). In addition, multiparous cows whose VWP was extended from 60 to 120 days had fewer days open after the end of the VWP, but for primiparous cows an extended VWP from 90 to 150 days did not affect reproductive performance (Arbel et al., 2001). Although an extended VWP may improve reproductive performance at the moment of insemination, possibly the farmer may decide that cows would get less time to become pregnant when the first insemination is delayed. This could increase culling due to fertility reasons (Niozas et al., 2019b). If farmers, however, deliberately aim for extended lactations, possibly they are willing to inseminate specific cows for a longer time.

2.3 Consequences for cow health and metabolism

An extended lactation implies a lower frequency of calvings in a cow's life. As calving is associated with an increased risk for disease, fewer calvings could improve cow health (Knight, 2005; Lehmann et al., 2014). In addition, fewer diseases could reduce risk for culling as a result of disease and increase the lifespan of cows. Limited information is available concerning disease incidence in an extended lactation. One study reported no differences in incidence of ketosis, milk fever, retained placenta, cystic ovaries, or displaced abomasum through 2 years for 108 cows with a VWP of 60 or 150 days (Van Amburgh et al., 1997). Another study used a simulation model to explore how extending the VWP from 50 to 150 days would change disease frequency during a four-year simulation, focusing on mastitis, metabolic disorders, and reproductive disorders (Allore and Erb, 2000). In that study, yearly rates of all diseases were lower when the VWP was extended. Cows with an extended VWP had a longer period of risk for mastitis as it was modeled to occur throughout the entire lactation. Nevertheless, primiparous cows with a VWP of 150 days had 15 % fewer mastitis cases per cow per year, and multiparous cows with a VWP of 150 days had 7 % fewer mastitis cases per cow per year, compared with primiparous and multiparous cows with a VWP of 50 days (Allore and Erb, 2000). As such, an extended lactation may be related with fewer calving-related disorders and fewer mastitis cases per year.

In addition, extending the lactation may reduce milk production before dry-off, which could limit the risk for udder infections during the dry period. One paper reported that around 25% of high-producing cows have a milk production above 20 kg/day in the final 30 days before dry-off (Green et al., 2008). Drying-off cows at a milk production of over 18 kg/d increased the risk for udder infections in the dry period and during the start of the next lactation (Rajala-Schultz et al., 2005; Odensten et al., 2007). For example, each increase in milk production at dry-off of 5 kg above 12.5 kg increased the chance for an intramammary infection at calving with 77 % (Rajala-Schultz et al., 2005). Cows with a VWP of 180 days had a lower milk production in the week of dry-off compared with cows with a VWP of 40 days (17.9 vs. 19.9 kg/d), and more cows were dried off at a milk production below 15 kg/d (54% vs. 34%; Niozas et al., 2019a). Cows with an average milk production below 15 kg/d in the final week before dry-off had a lower udder pressure in the first 9 days after dry-off compared with cows with an average milk production above 15 kg/d in the final week before dry-off (Bertulat et al., 2013). Therefore, an extended lactation might reduce udder pressure during the early phase of the dry period and herewith improve cow welfare.

In contrast, the reduced milk production at the end of the lactation also could be disadvantageous for the health status of dairy cows. First, a reduced milk production at the end of the lactation may be related with a rise in SCC (Österman et al., 2005; Niozas et al., 2019a). The greater SCC at the end of the extended lactation, however, has not been related with an increase in mastitis incidence (Sorensen et al., 2008; Niozas et al., 2019a), indicating that this rise in SCC was not the result of clinical mastitis. Possibly, the rise in SCC can be explained by the lower milk production or the decline in epithelial integrity at the end of the lactation (Sorensen et al., 2001).

Second, a reduced milk production at the end of the lactation may be related with an increased body condition at the end of the lactation (Niozas et al., 2019a). When body condition at dry-off was either < 3.5 or > 3.75 on a 5-point scale, feed intake after calving was greater for cows with the lower condition score (Schuh et al., 2019). Moreover, cows with a lower condition score of < 3.5 reached a positive energy balance at 10 weeks after calving, compared with cows with a greater condition score of > 3.75 who needed 12 weeks to return to a positive energy balance after calving (Schuh et al., 2019). In that study, cows with the higher condition score at dry-off had a greater plasma NEFA concentration after calving (Schuh et al., 2019), indicating more body fat mobilization and an increased risk for metabolic disorders (Morrow, 1976). In that case, even though the frequency of calvings is lower, cows with an extended lactation and an increased condition score at the end of the extended lactation may have an increased risk for metabolic disorders related with fat mobilization and a more severe NEB after the subsequent calving.

2.4 Consequences for farmer’s income

Several studies modeled the effect of extended lactations on the economic result (Holmann et al., 1984; Dijkhuizen et al., 1985; Schmidt, 1989; Strandberg and Oltenacu, 1989; Sørensen and Østergaard, 2003; Groenendaal et al., 2004; Inchaisri et al., 2011; Steeneveld and Hogeveen, 2012; Kok et al., 2019). In general, these studies concluded that a one-year CInt resulted in greater revenues, ranging between € 14 and € 110 per cow per year (Inchaisri et al., 2011; Steeneveld and Hogeveen, 2012; Kok et al., 2019). Lower revenues with an extended lactation could mainly be attributed to the relative high milk production in early lactation, when cows have their peak in milk, and the relative low milk production in late lactation (Strandberg and Oltenacu, 1989). The input data of these modeling studies, however, usually did not include lactations that were deliberately extended. In addition, cash flows depended on the length of the extension of the VWP, the parity of the cows, and factors such as the milk production level or the lactation persistency (Inchaisri et al., 2011; Steeneveld and Hogeveen, 2012; Kok et al., 2019).

It could be hypothesized that the effect of deliberate extended lactations on economic result is different from the existing modeling studies when: 1. Cows are blocked and randomly assigned to a certain VWP in an experimental setting where they are monitored for a complete lactation, and 2. Consequences for reproductive performance, feed intake, and disease incidence of individual cows are included in the economic evaluation. The effect of a deliberately extended lactation on economic result has been studied to a limited extent. Net returns per cow increased with $0.19 per day for primiparous cows or with $0.12 per day for multiparous cows when the VWP was extended with 60 days (Arbel et al., 2001) or with $0.75 per day when the VWP was extended from 60 to 150 days (Van Amburgh et al., 1997). These studies only included high-producing cows (Arbel et al., 2001) or stimulated milk production with the use of bST (Van Amburgh et al., 1997). This increase in net returns in extended lactations could indicate that an extended VWP may result in an improved economic result if cows are high-producing, or milk production is stimulated. Another study investigated the effect of extending the VWP from 60 to 88 d on cash flows (Stangaferro et al., 2018), and reported a numerical increase in yearly cash flow for primiparous cows, and a numerical decrease in yearly cash flow for multiparous cows. This limited extension of the VWP could still result in a one-year CInt, and as such it can be questioned if it is truly an extension of the lactation. However, it may indicate that for primiparous cows, an extended lactation can improve the economic result.

These experimental studies (Arbel et al., 2001; Van Amburgh et al., 1997; Stangaferro et al., 2018) show that cow characteristics such as milk production level or parity affect the economic consequence of an extended lactation, as was also indicated by earlier modeling studies (Inchaisri et al., 2011; Steeneveld and Hogeveen, 2012; Kok et al., 2019). In addition, feed supply and labor related with inseminations, disease treatments, and calves can be expected to be reduced when the CInt is extended deliberately. Furthermore, debate is ongoing how to handle the surplus calves born on dairy farms (LNV, 2021; Bolton and Von Keyserlingk, 2021). In this debate, one of the suggestions is that calves stay longer on the dairy farm, which would amplify the impact of extended lactations on costs for calf care and rearing.

2.5 Individual cow characteristics related with the response to an extended lactation

The effect of an extended lactation on milk characteristics depended on individual cow characteristics such as parity (Rehn et al., 2000; Lehmann et al., 2017), persistency (Sorensen et al., 2008, Pryce et al., 2010), milk production level (Mellado et al., 2016) and peak production (Lehmann et al., 2017). These cow characteristics could be used by farmers to select specific cows for an extended lactation, to limit the possible disadvantages of extended lactations such as a reduction in milk production or fattening at the end of the lactation. So far, cow characteristics to predict lactation performance in extended lactations were mostly based on milk characteristics in the current or previous lactation (Sorensen et al., 2008; Lehmann et al., 2017). Possibly, additional cow characteristics related with body condition or metabolism are also important in the decision to extend the lactation of an individual cow. For example, cows fed a total mixed ration (TMR) diet had a greater plasma glucose, insulin, and IGF-1 concentration between 332 and 612 DIM, and fewer of these cows fed a TMR diet were able to sustain the lactation for 670 days compared with grazing cows (Grainger et al., 2009; Delany et al., 2010). Therefore, it can be hypothesized that not only cow characteristics related with milk production level or parity, but also cow characteristics based on metabolism, energy partitioning, or hormonal status could affect the consequences of an extended lactation for individual cows.

3 Aim and outline of the thesis

The aim of this thesis was to evaluate the consequences of extended lactations for milk production, fertility, health, metabolism, and economic result. Moreover, the aim was to identify cow factors that determine the response of cows to an extended lactation. This way, lactation lengths may be customized for individual cows. As such, the objectives were to:

- Evaluate milk production and fertility on dairy farms that already apply extended lactations
- Asses the effect of a deliberately extended VWP on milk production characteristics, body weight development, metabolism, and economic results, for the current lactation and start of the next lactation
- Identify individual cow characteristics that can predict lactation performance in extended lactations

To study these objectives, a two-sided approach was applied: 1. A network of 14 Dutch dairy farmers that apply extended lactations on their farms was formed, to exchange knowledge and to evaluate data of commercial farms (farm data), and 2. A large animal experiment was conducted, where 154 cows were randomly assigned to a VWP of 50, 125, or 200 days, and followed for a complete lactation and the first 6 weeks of the next lactation (experimental data). To evaluate consequences of an extended lactation for milk production and fertility, data of the network of dairy farmers was used. This way, it could be investigated how extended lactations work out in practice when part of the cows is selected and managed for an extended lactation. Next to the farm data, the experimental data was used to evaluate consequences of an extended VWP for milk production, body weight, metabolism, and economic results. This way, cows could be blocked for certain characteristics and randomly assigned to a VWP, which made it possible to compare equal sets of cows in their response to a certain VWP. Moreover, individual cow characteristics could be investigated that may be related with this response.

The outline of the thesis is visualized in figure 3. In chapter 2, milk production characteristics and fertility characteristics are evaluated for cows with a different length of CInt and calving to first service interval (CFSI), of 13 farms where extended lactations are part of the management decisions of the farmer. In chapter 3, 4, and 5, the effect of an extended VWP on milk production characteristics, body condition, metabolism, and net partial cash flows is assessed, of cows that were randomly assigned to a VWP in a controlled experiment. Moreover, in chapter 3, 4, and 5, models are created to predict lactation performance of cows with different VWP, as a first step towards customized lactation lengths based on individual cow characteristics. In chapter 5, the prediction models are extended, and input is based on milk production and content, body condition, feed intake and energy balance, and plasma hormones and metabolites. The prediction models are based on the data of the experiment, but the motivations and strategies of the farmers of our network were an important input in the development of the factors for a successful extended lactation, i.e., the variables for lactation performance.

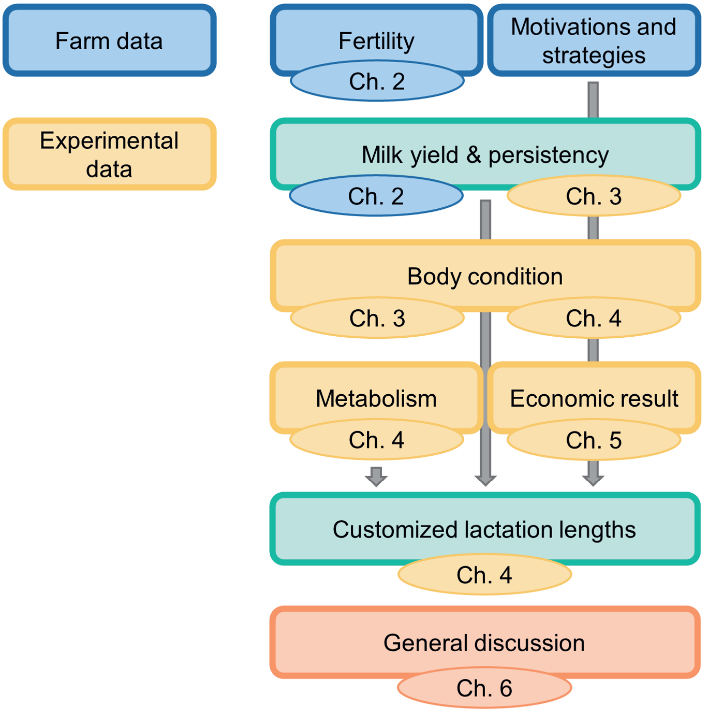


Figure 3. Topics that are covered in this thesis and according chapters. Blue chapters are based on farm data, yellow chapters are based on experimental data.

Chapter 2

Fertility and milk production on commercial dairy farms with customized lactation lengths

E.E.A. Burgers[1,2,†], A. Kok[1,†], R.M.A. Goselink[2], H. Hogeveen[3], B. Kemp[1], A.T.M. van Knegsel[1]

[1]Adaptation Physiology Group, Wageningen University & Research, Wageningen, the Netherlands
[2]Wageningen Livestock Research, Wageningen University & Research, Wageningen, the Netherlands
[3]Business Economics Group, Wageningen University & Research, Wageningen, the Netherlands
[†]These authors contributed equally to this work.

Journal of Dairy Science 104 (2021): 443 – 458

Abstract

Drying-off, calving, and start of lactation are critical transition events for a dairy cow. As a consequence, most animal health issues occur during these periods. By extending the voluntary waiting period for first insemination after calving, calving interval (CInt) can be extended, with possible positive effects for fertility and health. Some cows might be better suited for an extended CInt than others, due to differences in milk yield level, lactation persistency, or health status, which would justify a customized CInt based on individual cow characteristics. This study aims to investigate 13 farms with customized CInt, with respect to calving to first service interval (CFSI), accomplished CInt, services per conception (SC), conception rate at first artificial insemination (CR1AI), peak yield, lactation persistency, 305-d yield, and effective lactation yield. In total, 4,858 complete lactations of Holstein Friesian cows between 2014 and 2019 from the 13 farms were grouped by parity (1 or 2+) and CFSI (CFSI class; CFSI-1 < 84; 84 ≤ CFSI-2 < 140; 140 ≤ CFSI-3 < 196; 196 ≤ CFSI-4 < 252, CFSI-5 ≥ 252 d) or CInt (CInt class; CInt-1 < 364; 364 ≤ CInt-2 < 420; 420 ≤ CInt-3 < 476; 476 ≤ CInt-4 < 532, CInt-5 ≥ 532 d). Cow inseminations, available for 11 out of 13 farms (3,597 complete lactations), were grouped by parity (1 and 2+) and CFSI class or CInt class. The fertility and milk production characteristics were analyzed with generalized and general linear mixed models. The CFSI class was not associated with SC, but extended CInt class was associated with increased SC (CInt-1–5; 1.11–3.70 SC). More than 50% of cows in the CFSI class <84 d ended up in longer than expected CInt (>364 d), showing that these cows were not able to conceive for the desired CInt. More than 50% of cows in CInt classes 3 and higher (CInt ≥ 420 d) had an earlier first insemination before successful insemination (CFSI class 1; <196 d), showing that these extended CInt classes consisted of both cows with an extended waiting period for first insemination and cows that failed to conceive at earlier insemination(s). On most farms, lactation persistency was greatest in CInt class 1 (<364 d), probably related to the low peak yield in this class. When this shortest CInt class was excluded, persistency increased with extended CInt classes on most farms. Although at the majority of farms 305-d yield was greater in CInt ≥ 532 d, effective lactation yield at most farms was greatest in CInt from 364 to 531 d, especially for multiparous cows. Based on the results of this study, a CInt between 364 and 531 days seems most optimal for milk production, when high-yielding cows were selected.

Key words: extended calving interval, extended lactation, insemination, milk yield

1 Introduction

Drying-off, calving, and start of a new lactation are critical transition events for a dairy cow. Large changes in both physiology (e.g., calving, onset of milk production) and management (e.g., regrouping, start of milking) increase the risk for disease and culling (Butler, 2000; Fetrow et al., 2006; Pinedo et al., 2014). In most modern dairy systems, a cow faces these transition events every year, as a 1-yr calving interval (CInt) is usually aimed for. A 1-yr CInt is associated with a large average 305-d yield and better economic results compared with longer CInt (Strandberg and Oltenacu, 1989; Inchaisri et al., 2011; Steeneveld and Hogeveen, 2012).

It can be hypothesized that reducing the number of transition events per unit of time by extending CInt could be beneficial for fertility and health. First, insemination results could be improved because of a better metabolic status at the moment of insemination, as less inseminations are needed when insemination is delayed from 40 to 120 d after calving (Niozas et al., 2019b). Second, extending CInt could reduce the number of cows that are dried-off with a high milk yield (i.e., >18 kg/d), which could improve udder health (Rajala-Schultz et al., 2005; Odensten et al., 2007) and welfare (Zobel et al., 2015). Finally, some farmers aim to reduce the amount of calvings and calves born for the reduction of labor related to transition management and calf care, as well as the reduction in surplus of calves for cow replacement (Mohd Nor et al., 2014).

Although a 1-yr calving interval is usually aimed for due to maximal 305-d milk yield, milk losses due to an extended CInt could be less severe than reported in the modeling studies (Strandberg and Oltenacu, 1989; Inchaisri et al., 2011; Steeneveld and Hogeveen, 2012). First, these modeling studies were based on retrospective data, which implies a potential bias in the results because farmers likely tried to achieve a 1-yr CInt, with extended CInt indicating the involuntary consequence of health or fertility issues (Garverick et al., 2013; Carvalho et al., 2014). Second, some studies mainly reported 305-d yields. With an extended CInt, however, cows have a longer lactation period and less dry days per year, which influences both milk production per day and per year (Kok et al., 2019). Alternatively, milk production could be expressed as the total lactation including the dry period (i.e., averaged per day of CInt), similar to the effective lactation yield measure (Kok et al., 2016). Finally, the negative effect of pregnancy on milk yield might be delayed when CInt is extended, increasing lactation persistency (Bormann et al., 2002; Roche, 2003). Very persistent lactations could reduce milk losses, or possibly increase production, with an extended CInt (Arbel et al., 2001; Inchaisri et al., 2011; Kok et al., 2019).

Studies suggest that the optimal calving interval might be different for individual cows (Bertilsson et al., 1997; Kolver et al., 2007; Lehmann et al., 2017). Heifers had no or lower milk yield losses in increased CInt compared with older cows (Rehn et al., 2000; Österman and Bertilsson, 2003; Lehmann et al., 2016). In addition, milk yield level, body condition, or health status could be valuable cow characteristics that determine the response of cows to an extended voluntary waiting period for first insemination (Kolver et al., 2007; Lehmann et al., 2017). Recently, some farmers in the Netherlands started to customize CInt by extending the voluntary waiting period for first insemination after calving (VWP) for (part of) their herds. It is still a challenge for farmers, however, to select cows that have persistent lactations and therefore are capable of maintaining milk production with an extended CInt.

This study aimed to investigate farms with customized CInt with respect to calving to first service interval (CFSI), accomplished CInt, services per conception (SC), conception rate at first AI (CR1AI), peak yield, lactation persistency, 305-d yield, and effective lactation yield. We investigated multiannual data of 13 commercial Dutch dairy farms that managed their cows for a customized CInt, using various strategies to select individual cows for an extended VWP.

Table 1. Farm characteristics of 13 Dutch dairy farms in 2018 and their strategies to extend calving interval (CInt) by extending the voluntary waiting period for first insemination after calving (VWP)

Farm	Herd size	MS + freq[1]	Strategy to increase CInt				Mean CInt (d)	305-d yield (kg FPCM[2])
			VWP heifers[3]		VWP cows[3]			
A	110	CMS 2	FS[4]	“Dependent on days and production”	Fixed	80 d	453	9,342
B	120	CMS 2	Peak	30–34 kg: 100 d >34 kg: 150 d	Peak	40–45 kg: 100 d >45 kg: 150 d	411	10,596
C	75	CMS 2	Fixed	100 d	Fixed	100 d	468	8,625
D	65	AMS 2.7	Fixed, FS	150 d, “dependent on BCS and kg”	FS	“Dependent on cow and kg”	411	8,340
E	100	AMS 3.2	Level, FS	<30 kg, “in combination with days”	Level, FS	<30 kg, “in combination with days”	477	10,690
F	80	CMS 2	Fixed, Level	90 d, <30 kg	Fixed, Level	90 d, <40 kg	478	10,663
G	261	AMS 2.9	Peak	>30 kg: 100–150 d	Peak	>40 kg: 100 d	400	10,118
H	180	AMS 3	Fixed	110 d	Fixed	110 d	435	10,786
I	155	AMS 2.7	Level, FS	<30 kg, “in combination with condition”	Level	<33 kg	485	8,221
J	104	AMS 2.9	Level	<40 kg	Level	<45 kg	423	10,359
K	120	CMS 2	Level	<28 kg	Level	<38 kg	462	9,854
L	75	CMS 2	Fixed, FS	3–6 mo, “dependent on lactation value and condition”	Fixed, FS	3–9 mo, “dependent on lactation value and condition”	473	8,579
M	50	AMS 3	FS	“Dependent on production and condition”	FS	“Dependent on production and condition”	421	9,048

[1] MS = milking system; freq = milking frequency; CMS = conventional milking system; AMS = automatic milking system. For AMS, average frequency of milking is shown, as milking frequency can vary between cows and throughout lactation.

[2] FPCM = fat- and protein-corrected milk.

[3] Indicates in the case of farmer’s statement what the VWP is based on; in the case of a fixed VWP, the waiting period in days, and in the case of level, the level of milk yield below which insemination is started.

[4] FS = farmer’s statement. Not all farmers have a clear cut-off value to start insemination; some look at multiple factors such as BCS, health, and other cow characteristics.

2 Materials and methods

2.1 Herds

With an advertisement in a Dutch farmers magazine in 2017, farmers were asked to join a network group concerning the practical applications and implications of extended CInt on farms. This advertisement resulted in 13 Dutch dairy farmers with Holstein Friesian cows that deliberately extend the VWP for (part of) their cows and that were willing to share their milk production data. From these 13 farmers, 11 farmers were able to share their insemination data. Herd size, milking system, and average milking frequency, as well as the individual strategies to increase VWP and the accomplished mean CInt, are presented in Table 1. Criteria to select cows for an extended VWP differed among farmers. Some farmers used a fixed extended VWP for all cows, meaning that they waited a certain number of days after calving before starting insemination. Other farmers selected individual cows for an extended VWP based on daily milk yield, meaning that they waited until milk production dropped below a certain level before starting insemination. A few farmers selected cows for an extended VWP based on their peak yield; a greater peak yield implied a longer waiting period before start of insemination.

2.2 Data

Data of the 13 farms were retrieved via the Dutch milk recording system (CRV, Arnhem, the Netherlands). Only data from complete lactations with a known CInt (defined as the period from calving date to next calving date) were used. Cow lactations were grouped by parity (1 or 2+) and CFSI (CFSI-1 < 84; $84 \leq$ CFSI-2 < 140; $140 \leq$ CFSI-3 < 196; $196 \leq$ CFSI-4 < 252; CFSI-5 ≥ 252 d) or CInt class (CInt-1 < 364; $364 \leq$ CInt-2 < 420; $420 \leq$ CInt-3 < 476; $476 \leq$ CInt-4 < 532; CInt-5 ≥ 532 d), where each next CInt class was an extension of CInt with 8 wk. The CFSI classes were designed to match the CInt classes based on an assumed gestation length of 280 d and conception at first insemination. The Appendix gives an overview of the number of lactations per parity class, per CInt class, per farm (Appendix Table A1), and the mean CInt per parity class, per CInt class, per farm (Appendix Table A2). The Appendix also gives an overview of the number of lactations per parity class, per CFSI class, and per farm (Appendix Table A3), and the mean CFSI per parity class, per CFSI class, and per farm (Appendix Table A4).

Insemination data

Insemination data were available for 11 out of 13 farms, from February 2013 until March 2019. The original data set consisted of 5,487 lactations. In total, 1,890 incomplete lactations were removed. The final data set for analysis of inseminations included only complete lactations with insemination data available and consisted of 3,597 complete lactations with 6,968 inseminations. Cow inseminations were grouped by parity (1 or 2+) and CFSI or CInt class.

Milk production data

Milk production data were available for all 13 farms. Milk yield and composition were recorded every 4 to 6 wk, from January 2014 until January 2019. The original data set consisted of 8,447 lactations. In total, 3,589 incomplete lactations were removed. From these incomplete lactations, 1,499 lactations started after January 2018, and therefore these are likely to be ongoing at the end of the data set. The final data set for analysis of milk production consisted of 4,858 complete lactations with 43,859 milk records. Milk yield was converted to fat- and protein-corrected milk (FPCM) as:

$$\mathrm{FPCM(kg)} = \mathrm{milk\ (kg)} \times (0.337 + 0.116 \times \mathrm{fat\ (\%)} + 0.06 \times \mathrm{protein\ (\%)})$$ (CVB, 2012).

2.3 Statistical analysis

Insemination data

Number of SC were analyzed per CFSI classes and per CInt classes to compare between aimed CInt and result in practice. Insemination data were analyzed using 4 models. The SC was not normally distributed but followed a Poisson distribution. Number of SC per CFSI class was analyzed using a generalized linear mixed model with a Poisson distribution in SAS (PROC GLIMMIX, version 9.4, SAS Institute Inc., Cary, NC; model 1). The final model included fixed effects of parity, farm, and CFSI class. There were no interaction effects; these were removed from the model by backward selection.

The CR1AI per CFSI class was analyzed using a generalized linear mixed model with a binary distribution in SAS (PROC GLIMMIX; model 2). The final model included fixed effects of parity, farm, and CFSI class. This procedure modeled the probability that there was conception after first insemination.

The number of SC per CInt class was analyzed using a generalized linear mixed model with a Poisson distribution in SAS (PROC GLIMMIX; model 3). The final model included fixed effects of parity, farm, and CInt class. The interaction of CInt class × farm was retained in the model by backward selection of interaction effects.

The CR1AI per CInt class was analyzed using a generalized linear mixed model with a binary distribution in SAS (PROC GLIMMIX; model 4). The final model included fixed effects of parity, farm, and CInt class. All *P*-values of the least squares means were adjusted with a Bonferroni adjustment.

Milk production data

Milk production results were analyzed per CFSI class and per CInt class, with a mixed model in SAS (PROC MIXED), using 4 models. Two models allowed fixed effects of CFSI class or CInt class, parity, and lactation curve parameters (models 5 and 6). Significant interactions ($P < 0.05$) between the fixed effects and the lactation curve parameters were retained in the final model by backward selection. In these models, cow lactation was added as a random effect nested within farm.

Next, farm was added as a fixed effect to both the CFSI class model and the CInt class model (models 7 and 8). The results for the CFSI class model per farm can be found in Supplemental tables S1–S4. Significant interactions ($P < 0.05$) between the fixed effects and the lactation curve parameters were retained in the final model by backward selection. In addition to the fixed effects, the model included a random effect for repeated measures per cow lactation, assuming an unstructured covariance-structure. Based on these models, lactation curves were fitted using a Wilmink curve extended with a linear negative effect of gestation on milk production, starting at a fixed delay after conception (Wilmink, 1987; Strandberg and Lundberg, 1991):

$$y_t = a + b_t + c^{(-k+1)} + b_{gest} \times \max\left(\left(D_{gest} - D_{delay}\right), 0\right),$$

where a, b_t, c, and k (assumed at 0.05) represent the shape of the Wilmink lactation curve (Wilmink, 1987) and b_{gest} represents the linear negative effect of days in gestation (D_{gest}) from a fixed delay (D_{delay}) after conception (Strandberg and Lundberg, 1991). The D_{delay} was determined for the entire data set; the best fit, based on the Bayesian information criterion, was found for a gestation effect starting at 161 d after conception. From the lactation curves, we derived peak yield, lactation persistency, 305-d yield, and effective lactation yield. Lactation persistency was defined as the slope of the lactation curve from d 100 until d 212 in lactation in kilograms per day. The 305-d yield was calculated as the area under the curve in the first 305 days of lactation. Effective lactation yield was calculated as the total milk yield from calving to next calving (including the dry period) and expressed as FPCM per day of CInt (Kok et al., 2016; Lehmann et al., 2016), using the average CInt for each CInt class × parity class × farm combination, and assuming a 6-wk dry period.

3 Results

3.1 Calving to first service interval and calving interval

Based on their CFSI, cows had an expected CInt, when assuming conception at first insemination. Cows did not always end up in the expected CInt classes (Table 2). From the cows in the CFSI-1 class (<84 d), almost 50% ended up in the corresponding CInt class (i.e., CInt-1; <364 d). With extending CFSI class, this percentage increased. Per CFSI class, 50.2, 36.1, 36.0, and 28.9% of cows (for CFSI-1, CFSI-2, CFSI-3, CFSI-4, respectively) ended up in a higher CInt class than planned for, based on first insemination. A small proportion of cows ended up in a shorter CInt than expected from first insemination due to a gestation shorter than 280 d.

Based on their CInt, cows had an expected CFSI, when assuming conception at first insemination. Cows did not always originate in these expected CFSI classes (Table 3). More than 50% of cows in CInt classes 3 and higher (CInt ≥ 420 d) had an earlier first insemination before successful insemination (CFSI < 196 d; Table 3).

Table 2. Percentage of cows per calving to first service interval (CFSI) class per calving interval (CInt) class (total lactations = 3,597)

CInt class[1] (n of lactations)	CFSI class[2] (n of lactations)				
	CFSI-1 (939)	CFSI-2 (1,736)	CFSI-3 (569)	CFSI-4 (211)	CFSI-5 (142)
CInt-1 (509)	49.8[3]	2.4	—	—	—
CInt-2 (1,359)	28.2	61.5	4.6	—	—
CInt-3 (813)	12.3	20.3	59.4	3.3	—
CInt-4 (471)	5.5	9.0	20.4	67.8	2.8
CInt-5 (445)	4.2	6.8	15.6	28.9	97.2
Total	100	100	100	100	100

[1] CInt class: CInt-1 <364; 364 ≤ CInt-2 <420; 420 ≤ CInt-3 <476; 476 ≤ CInt-4 <532; CInt-5 ≥532 d.
[2] CFSI class: CFSI-1 <84; 84 ≤ CFSI-2 <140; 140 ≤ CFSI-3 <196; 196 ≤ CFSI-4 <252; CFSI-5 ≥252 d.
[3] Values on the diagonal indicate the percentage of cows that end up in planned CInt class.

Table 3. Percentage of cows per calving interval (CInt) class per calving to first service interval (CFSI) class (total lactation = 3,597)

CFSI class[1] (n of lactations)	CInt class[2] (n of lactations)				
	CInt-1 (509)	CInt-2 (1,359)	CInt-3 (813)	CInt-4 (471)	CInt-5 (445)
CFSI-1 (939)	91.9[3]	19.5	14.2	11.0	8.8
CFSI-2 (1,736)	8.1	78.6	43.4	33.1	26.5
CFSI-3 (569)	—	1.9	41.6	24.6	20.0
CFSI-4 (211)	—	—	0.9	30.4	13.7
CFSI-5 (142)	—	—	—	0.9	31.0
Total	100	100	100	100	100

[1] CFSI class: CFSI-1 <84; 84 ≤ CFSI-2 <140; 140 ≤ CFSI-3 <196; 196 ≤ CFSI-4 <252; CFSI-5 ≥252 d.
[2] CInt class: CInt-1 <364; 364 ≤ CInt-2 <420; 420 ≤ CInt-3 <476; 476 ≤ CInt-4 <532; CInt-5 ≥532 d.
[3] Values on the diagonal indicate the percentage of cows that originate from expected CFSI class.

3.2 Services per conception and conception rate at first AI

The number of SC ranged from 1 to 12 (Table 4). The number of SC was equal for extending CFSI classes. Parity class, farm, and CFSI class all affected CR1AI. The CR1AI was lower for CFSI class 3 compared with CFSI class 2. The number of SC increased with CInt classes (Table 5) and differed among farms. Both farm and CInt class affected CR1AI. The CR1AI decreased with extending CInt class. In CInt class 1, there were some farms without cows that needed multiple inseminations. In CInt class 5, some farms had zero cows that conceived after 1 insemination.

Table 4. Services per conception (SC) and conception rate at first insemination per calving to first service interval (CFSI) class

	CFSI class (d)				
Item	CFSI-1 (<84)	CFSI-2 (84–139)	CFSI-3 (140–195)	CFSI-4 (196–251)	CFSI-5 (≥252)
Lactations (n)	939	1,736	569	211	142
Services per conception (mean ± SE)	1.90 ± 0.05	1.82 ± 0.04	1.93 ± 0.06	1.76 ± 0.10	1.89 ± 0.13
Range SC[1] (minimum–maximum)	1–11	1–12	1–9	1–8	1–8
Conception rate[2] (mean %)	52.8^{ab}	57.3^{a}	49.5^{b}	57.3^{ab}	51.7^{ab}

^{a,b}Means within a row with different superscripts differ (*P* = 0.03).
[1] Range SC is presented per cow per lactation.
[2] Conception rate is defined as the percentage of cows pregnant at first AI.

Table 5. Services per conception (SC) and conception rate at first insemination per calving interval (CInt) class

	CInt class (d)				
Item	CInt-1 (<364)	CInt-2 (364–419)	CInt-3 (420–475)	CInt-4 (476–531)	CInt-5 (≥532)
Lactations (n)	509	1,359	813	471	445
Services per conception (mean ± SE)	1.11^{a} ± 0.13	1.33^{a} ± 0.04	1.94^{b} ± 0.06	2.62^{c} ± 0.09	3.70^{d} ± 0.11
Range SC[1] (minimum–maximum)	1–4	1–5	1–7	1–7	1–12
Conception rate[2] (mean %)	99.7^{abc}	74.0^{a}	36.4^{b}	17.4^{c}	0.28^{abc}

$^{a-d}$Means within a row with different superscripts differ (*P* < 0.0001).
[1] Range SC is presented per cow per lactation.
[2] Conception rate is defined as the percentage of cows pregnant at first AI.

3.3 Lactation curves

Peak yield, lactation persistency, 305-d yield, and effective lactation yield were associated with parity class, CFSI class (Table 6), and CInt class (Table 7). For parity 1, peak yield was highest in CFSI-4 and CInt-4. For parity 2+, peak yield was highest in CFSI-4 and CInt-5. For both parity classes, persistency was highest in CFSI-5 and CInt-5. For parity 1, 305-d yield was highest in CFSI-4 and CInt-4, as was the effective lactation yield. For parity 2+, 305-d yield was highest in CFSI-4 and CInt-5; however, effective lactation yield was highest in CFSI-2 and CInt-2.

Moreover, peak yield, persistency, 305-d yield, and effective lactation yield were associated with a farm effect and interactions with the lactation curve characteristics (Appendix Table A5).

Table 6. Peak yield, lactation persistency, 305-d yield, and effective lactation yield per calving to first service interval (CFSI) class per parity class

		CFSI class (d)				
Item	Parity class	CFSI-1 (<84)	CFSI-2 (84–139)	CFSI-3 (140–195)	CFSI-4 (196–251)	CFSI-5 (≥252)
Peak yield (kg of FPCM[1])	1	31.0	31.9	31.3	32.9	32.2
	2+	40.2	43.2	43.4	44.8	43.4
Lactation persistency (kg of FPCM per day)	1	−0.027	−0.029	−0.023	−0.023	−0.020
	2+	−0.062	−0.064	−0.059	−0.058	−0.055
305-d yield (kg of FPCM first 305 d)	1	8,641	8,805	8,803	9,262	9,167
	2+	10,325	11,095	11,205	11,557	11,303
Effective lactation yield (kg of FPCM per day of calving interval)	1	25.2	25.6	25.5	26.4	25.9
	2+	29.5	31.3	30.6	30.5	28.3

[1]FPCM = fat- and protein-corrected milk.

Table 7. Peak yield, lactation persistency, 305-d yield, and effective lactation yield per calving interval (CInt) class per parity class

		CInt class (d)				
Item	Parity class	CInt-1 (<364)	CInt-2 (364–419)	CInt-3 (420–475)	CInt-4 (476–531)	CInt-5 (≥532)
Peak yield (kg of FPCM[1])	1	30.1	31.7	32.1	32.1	31.1
	2+	39.5	42.3	43.0	43.4	43.6
Lactation persistency (kg of FPCM per day)	1	−0.025	−0.029	−0.027	−0.025	−0.020
	2+	−0.060	−0.063	−0.062	−0.059	−0.054
305-d yield (kg of FPCM first 305 d)	1	8,577	8,797	8,907	9,019	8,844
	2+	10,384	10,870	11,035	11,223	11,440
Effective lactation yield (kg of FPCM per day of calving interval)	1	24.7	25.7	25.8	25.9	25.2
	2+	29.9	31.1	30.6	30.3	29.1

[1]FPCM = fat- and protein-corrected milk.

3.4 Peak yield and persistency per farm

Effects of CInt class on peak yield and lactation persistency were dependent on parity and farm. The modeled peak yield per farm ranged from 20.3 to 37.6 kg/d of FPCM for parity 1, and from 26.8 to 51.4 kg/d of FPCM for parity 2+ (Supplemental table S5). For 9 out of 13 farms in parity 1, peak yield was lowest for CInt-1 compared with the peak yield of the other CInt classes within farms. For the other 4 farms, peak yield was lowest for CInt-2 (farm L), CInt-3 (farms C and M), and CInt-5 (farm J; Figure 1A). For parity 2+, the peak yield was lowest for CInt-1 for all farms compared with the peak yield of the other CInt classes within farms (Figure 1B).

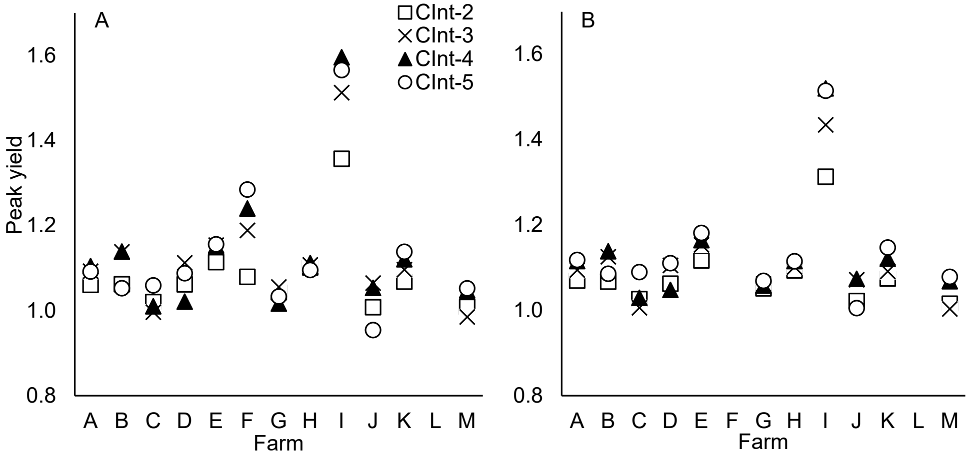


Figure 1. Peak yield per farm (A–M) per calving interval (CInt) class relative to CInt-1 (CInt-1 = 1) for cows with parity 1 (A) or parity 2+ (B). The CInt classes are CInt-1 (<364 d), CInt-2 (364–419 d), CInt-3 (420–475 d), CInt-4 (476–531 d), and CInt-5 (≥532 d). Farm L parity 1 could not be computed. Farm F and farm L do not have lactations with parity 2+ in CInt-1 and are therefore not shown.

The lactation persistency per farm ranged from 0.003 to 0.052 kg of FPCM reduction per day for parity 1, and from 0.009 to 0.102 kg of FPCM reduction per day for parity 2+ (Supplemental table S6). For 7 out of 13 farms in parity 1, lactation persistency was greatest for CInt-5 compared with the lactation persistency of the other CInt classes within farms. For the other 6 farms, lactation persistency was greatest for CInt-1 (farms B, E, I, and K), and CInt-4 (farms C and L; Figure 2A). For 7 out of 13 farms in parity 2+, lactation persistency was greatest for CInt-5 compared with the lactation persistency of the other CInt classes within farms. For the other 6 farms, lactation persistency was greatest for CInt-1 (farms B, E, I, K, and L), and CInt-4 (farm C; Figure 2B).

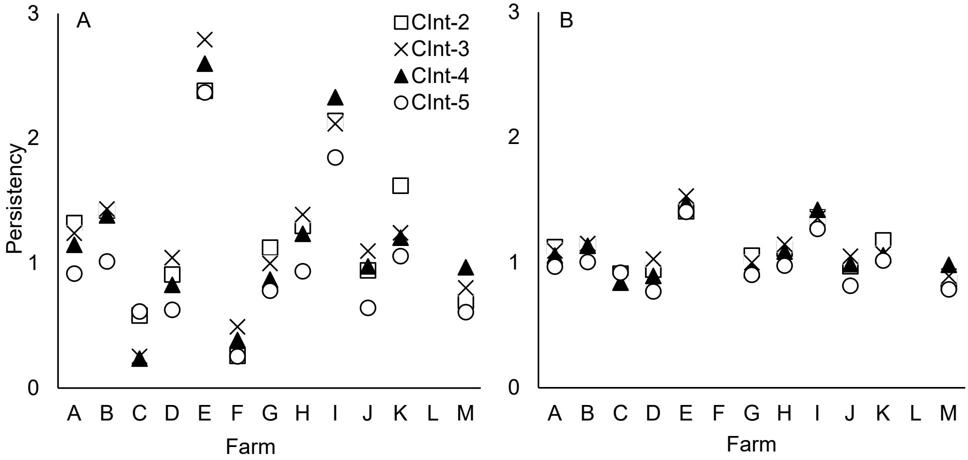


Figure 2. Persistency per farm (A–M) per calving interval (CInt) class relative to CInt-1 (CInt-1 = 1) for cows with parity 1 (A) or parity 2+ (B). The CInt classes are CInt-1 (<364 d), CInt-2 (364–419 d), CInt-3 (420–475 d), CInt-4 (476–531 d), and CInt-5 (≥532 d). Farm L parity 1 could not be computed. Farm F and farm L do not have lactations with parity 2+ in CInt-1 and are therefore not shown.

3.5 The 305-d yield and effective lactation yield per farm

Effect of CInt class on 305-d yield and effective lactation yield depended on parity and farm. The 305-d yield per farm ranged from 5,822 to 10,843 kg of FPCM for parity 1, and from 6,867 to 13,546 kg of FPCM for parity 2+ (Supplemental table S7). For parity 1, 9 out of 13 farms had greatest 305-d yield for CInt-5 compared with the other CInt classes within farms. For the other 4 farms, 305-d yield was greatest for CInt-3 (farms G and J) and CInt-4 (farms B and L; Figure 3A). For 10 out of 13 farms, 305-d yield was lowest for CInt-1, and for 3 farms 305-d yield was lowest for CInt-3 (farms C and M) and CInt-5 (farm J). For 11 out of 13 farms in parity 2+, 305-d yield was greatest for CInt-5 compared with the other CInt classes within farms. The other 2 farms had greatest 305-d yield for CInt-4 (farms B and J). Except for farm M, all farms had lowest 305-d yield for CInt-1 (Figure 3B).

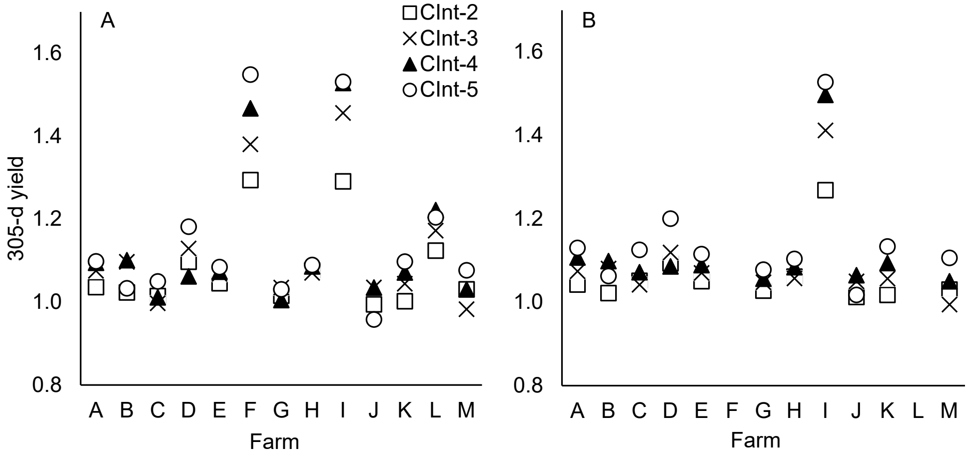


Figure 3. The 305-d yield per farm (A–M) per calving interval (CInt) class relative to CInt-1 (CInt-1 = 1) for cows with parity 1 (A) or parity 2+ (B). The CInt classes are CInt-1 (<364 d), CInt-2 (364–419 d), CInt-3 (420–475 d), CInt-4 (476–531 d), and CInt-5 (≥532 d). Farm F and farm L do not have lactations with parity 2+ in CInt-1 and are therefore not shown.

The effective lactation yield per farm ranged from 16.7 to 32.6 kg of FPCM per day for parity 1, and from 19.8 to 35.5 kg of FPCM per day for parity 2+ (Supplemental table S8). For parity 1, 6 out of 13 farms had greatest effective lactation yield for CInt-5 compared with the other CInt classes within farms. For the other 7 farms, effective lactation yield was greatest for CInt-2 (farm H), CInt-3 (farms G and J), and CInt-4 (farms B, E, I, and L; Figure 4A). For parity 2+, 6 out of 13 farms had greatest effective lactation yield for CInt-2 compared with the other CInt classes within farms. For the other 7 farms, effective lactation yield was greatest for CInt-3 (farms B and J), CInt-4 (farms A, I, K, and L), and CInt-5 (farm F; Figure 4B).

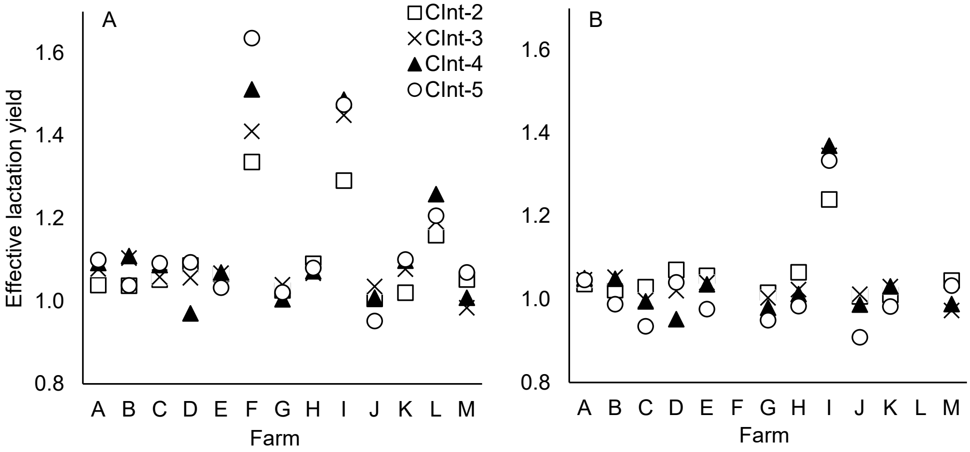


Figure 4. Effective lactation yield per farm (A–M) per calving interval (CInt) class relative to CInt-1 (CInt-1 = 1) for cows with parity 1 (A) or parity 2+ (B). The CInt classes are CInt-1 (<364 d), CInt-2 (364–419 d), CInt-3 (420–475 d), CInt-4 (476–531 d), and CInt-5 (≥532 d). Farm F and farm L do not have lactations with parity 2+ in CInt-1 and are therefore not shown.

4 Discussion

The current study aimed to investigate fertility and milk production on farms that customize CInt of their cows. On these farms, different cow characteristics were used to determine which cows to extend CInt. These cow characteristics could differ between farms and between years. This study adds insight to the consequences of customized lactation management in practice.

4.1 Fertility

Calving interval was extended by extending the VWP. Calving to first service interval was used as a measure of the VWP. In the current study, CFSI class was not related to SC. The CFSI class 3 (140–195 d) had a lower CR1AI compared with CFSI class 2 (84–139 d), however there were no differences with or between the other CFSI classes. Earlier studies found a decrease in SC (Larsson and Berglund, 2000; Niozas et al., 2019b) and an improved CR1AI (Larsson and Berglund, 2000; Inchaisri et al., 2011; Niozas et al., 2019b) when VWP was extended. This was explained by the delay of insemination until a cow is possibly out of the negative energy balance (NEB). The NEB in early lactation has been associated with impaired fertility, as the lack of glucose and increased free fatty acid concentration may impair oocyte quality (Jorritsma et al., 2004; Leroy et al., 2006; Fouladi-Nashta et al., 2007). Possibly, in this study, CFSI was specifically extended for cows with high milk production as a result of farmers' strategies. Several farmers aim at insemination at a specific milk production level for all cows. This could have resulted in a similar metabolic status, and with that a similar health status at time of conception (Butler et al., 1981). A similar health status could mean similar fertility, leading to similar success of insemination (i.e., SC and conception rate at first AI; Niozas et al., 2019b). The current study, moreover, used retrospective farm data. It is unknown whether an extended CFSI was a deliberate decision of a farmer, a real measure of the VWP, or the result of a cow not displaying estrus. Therefore, the extended CFSI classes could consist of both cows that are deliberately inseminated later and cows with estrus or health problems that could not be inseminated earlier. In extended CFSI classes, the maximum SC decreased. This finding might be related to improved fertility. With extending CFSI classes, moreover, the percentage of cows that ended up in higher CInt classes decreased, which might imply improved fertility after delayed insemination. Alternatively, cows with an extended CFSI may get fewer chances to conceive before they are replaced because a lower milk yield at that time might make it undesirable to extend CInt further. The present study did not include incomplete lactations, which may have skewed SC if cows were allowed fewer inseminations in the case of an extended VWP.

The farmers in the current study aimed to customize CInt by extending VWP. An increased CFSI, however, was not always the reason for an extended CInt. In fact, around 70% of cows in a CInt ≥ 476 d (CInt-4 and CInt-5) were first inseminated aiming at a shorter CInt. From CInt-3 onwards (≥420 d), the majority of cows had a lower CFSI (CFSI < 196 d) than expected based on CInt class. As a consequence, the extended CInt classes consisted of both cows with a delayed first insemination (either voluntarily or due to lack of estrus) and cows that were unable to conceive earlier and therefore needed multiple inseminations to become pregnant. Overall SC in this study was 1.94; SC was 1.90 for cows in parity 1 and 1.96 for cows in parity 2+, with a maximum of 12 inseminations per cow. Farmers in this study may have been more accepting toward an extended CInt, and therefore were more inclined to inseminate a cow with difficulties to conceive multiple times, rather than replacing that cow, compared with farmers that aim for a 1-yr CInt.

Cows could end up in extended CInt due to poor fertility and therefore more days to pregnancy than aimed for based on first insemination. In fact, 50.2% of cows with CFSI < 84 d ended up in longer than expected CInt (>364 d). This showed that these cows were not able to conceive for the desired CInt, and moreover, that the longer CInt classes consisted of both cows selected for an extended CInt and cows unable to conceive sooner. A 1-yr CInt is still generally advised for an optimal economic result (Holmann et al., 1984; Steeneveld and Hogeveen, 2012), and therefore it can be assumed that the majority of Dutch dairy farmers aim for a 1-yr CInt. Less than 2% of farmers in the Netherlands, however, achieve an average CInt of <369 d (CRV, 2019).

4.2 Lactation curves

In the current study, peak yield and lactation persistency were calculated according to fitted lactation curves. Earlier studies pointed out that, in terms of milk production, extending CInt seemed more successful for cows with greater lactation persistency (Arbel et al., 2001; Inchaisri et al., 2011; Kok et al., 2019), or cows with greater peak yield (Rehn et al., 2000; Lehmann et al., 2017). In the current study, the peak yield was lower for CFSI-1 class (<84 d) and for CInt-

1 class (<364 d) compared with the peak yield of the other CFSI and CInt classes. Low peak yield in the short CFSI CInt classes could be related to 2 aspects. First, in the present study, it could reflect the strategy of farmers to start insemination soon when peak yield is low. Not all cows in extended CInt, however, had an extended CInt because of delayed insemination. Second, cows with a low peak yield may resume ovarian cyclicity earlier (Opsomer et al., 1998; Shrestha et al., 2004) and express estrus more easily, as milk yield has been found to negatively correlate with estrus expression (Lopez et al., 2004; Holman et al., 2011; Cutullic et al., 2012). Cows with a low peak yield, moreover, were more likely to conceive in 1 or 2 inseminations (Lean et al., 1989).

Persistency was greater in the longer CFSI classes, possibly reflecting successful selection of cows with greater persistency for extended CFSI. Farmers had different strategies to select cows for extended CFSI. Farmers that based their strategy on production level indirectly took persistency into account, as more persistent cows will take longer to drop below the cut-off level for milk yield and are thus inseminated later and end up in greater CFSI classes. Despite that many cows in the extended CInt classes did not originate from an extended CFSI, lactation persistency increased from CInt-2 to CInt-5 (364–532 d). There are a few possible reasons for this positive relationship between CInt length and persistency. First, possibly only high-producing, persistent cows were given many chances to become pregnant, resulting in a higher proportion of persistent lactations in an extended CInt, and low-producing or less persistent cows may have been culled and therefore did not end up in the data set. Second, it is possible that mainly high-producing cows had more difficulties to conceive and as a result involuntarily ended up in extended CInt (Chebel et al., 2004; Walsh et al., 2011). Third, increased lactation persistency in extended CInt has been related to a delayed effect of pregnancy on the lactation curve (Brotherstone et al., 2004). The persistency in the present study was calculated between d 100 and 212, a time that pregnancy was assumed to not yet affect the lactation curve (Strandberg and Lundberg, 1991; Penasa et al., 2016). However, gestation may reduce milk yield from the first month onward, which could already have reduced persistency between d 100 and 212 in lactation (Olori et al., 1997). The greater lactation persistency in the longer CInt classes could thus be related to selection of persistent animals for a long CInt or cow physiology (i.e., poor fertility of high-producing cows or a delayed pregnancy effect after later insemination; Olori et al., 1997; Brotherstone et al., 2004; Chebel et al., 2004).

On some farms, the greatest lactation persistency was found for CInt-1 (<364 d). For these farms, however, the lowest peak yield was also found for CInt-1. A low peak yield is related to high persistency, whereas a high peak yield is related to low persistency (Dekkers et al., 1998). A high peak was related to a more severe NEB, and a more severe NEB has been related to an altered metabolic status, associated with increased plasma non-esterified fatty acid concentration and a greater incidence of metabolic diseases (Esposito et al., 2014). Both elevated levels in plasma of free fatty acids and β-hydroxybutyrate in early lactation (Chen et al., 2016) and the occurrence of mastitis after peak yield (Appuhamy et al., 2007) have been related to reduced lactation persistency. When peak yield was delayed and lowered as a consequence of metabolic diseases in early lactation (i.e., before peak yield), persistency was found to increase, indicating that a low peak is related to an increased persistency (Appuhamy et al., 2007; Hostens et al., 2012).

The 305-d yield reflects the lactation potential of a dairy cow (Kuhn and Hutchison, 2005; Kok et al., 2016), and is therefore expected to be correlated with effective lactation yield. For cows in parity 1, both 305-d yield and effective lactation yield were greatest in CFSI-4 (196–251 d) and in CInt-4 (476–531 d). For cows in parity 2+, however, greatest 305-d yield was found in CFSI-4 (196–251 d) and in CInt-5 (≥532 d), whereas greatest effective lactation yield was found in CFSI-2 (84–139) and in CInt-2 (364–419 d). Within farms, overall greatest 305-d yield was found in CInt-5 (parity 1: 9 out of 13 farms; parity 2+: 11 out of 13 farms), however, they mostly did not realize the greatest effective lactation yield in CInt-5. For cows with parity 1, greatest effective lactation yield was found in CInt-5 in 6 farms. By selecting the best cows in terms of 305-d yield for extended CInt, it was thus possible to realize high effective lactation yield of first parity cows in extended CInt. In the other cases, effective lactation yield was still comparable to the effective lactation yield in the other CInt classes, and greater than the effective lactation yield in CInt-1. For cows with parity 2+, greatest effective lactation yield was found in CInt-5 in 1 farm. In the other farms where 305-d yield was greatest in CInt-5, effective lactation yield was often lowest in CInt-5. The effective lactation yield corrects for CInt (Kok et al., 2016). Thus, when corrected for length of CInt, the best cows in terms of 305-d yield in the CInt-5 group did not have the greatest milk production per day in extended CInt. This can be explained by a decrease in their milk production toward the end of their long lactation, and these cows would probably have accomplished greater effective lactation yield in shorter CInt (Kok et al., 2019).

The greatest 305-d yield found in CInt-5 is probably partly due to selection of cows with higher milk yield for a longer CInt by using peak yield or daily milk level to determine VWP (7 farmers). Moreover, some farmers gave their cows many chances to become pregnant (up to 12 inseminations). Depending on parity and milk yield, cows can be inseminated up to 16 mo in milk before it becomes more profitable to replace that cow (Inchaisri et al., 2012). In the current study, number of inseminations per pregnancy was much greater in the extended CInt classes compared with the shorter CInt. If only cows in CInt-5 that were planned for a shorter CInt were considered, average number of inseminations for the CInt-5 group was 4.33. Therefore, in the current study, some of the cows in CInt-5 may have been there because these were high-producing cows that were either selected for a long CInt or were unable to conceive early and ended up in extended CInt due to multiple inseminations, contributing to the high 305-d yield in this group.

Farmers that have a fixed VWP in days for all their cows do not take individual milk yield or lactation persistency into account when assigning cows to an extended CInt. As a result, some of these farmers did not realize more persistent lactations in extended CInt. In extended CInt it is especially important to have more persistent lactations to minimize losses from extending CInt (Kok et al., 2019). Some of these farmers with a fixed VWP in days, however, argued that their goal is not to maximize milk yield per cow, but to identify and select cows capable of maintaining lactation in an extended CInt. When having the same VWP for all cows, a farmer can use cow performance in an extended VWP strategy to select cows suitable for extended CInt for the next generation.

There was a large variation in lactation curve characteristics among farms. The main reason for this was probably due to a large variation in management or possibly genetics among farms in general. When looking within farms, however, similar patterns were found among the farms for the different CInt and CFSI classes concerning milk yield and lactation curve characteristics (Figures 1–4; Supplemental tables S1–8). Although absolute values among farms differ, cows with a higher peak, higher 305-d milk yield, and higher persistency still ended up in longer CInt classes on most farms.

4.3 Motivation to extend the calving interval

Instead of maximizing FPCM yield, farmers in the current study were interested in customizing CInt for other reasons. First, farmers aimed for potential health benefits related to an extended CInt. Extending CInt increased the time between critical transition events and could lower the number of cows that are being dried-off at high milk yields, therewith possibly improving health

(Knight, 2005; Lehmann et al., 2014; Niozas et al., 2019a). Second, some farmers aimed for fewer calves born. At farm level, fewer calves result in less income from calves sold, but because calf prices are limited, it might be a benefit due to a reduction in costs (Mohd Nor et al., 2012). On a typical dairy farm, replacement rate can be assumed to be around 30% (Mohd Nor et al., 2014), indicating a surplus of calves that need care, labor, and feed when a 1-yr CInt is applied. A problem with selecting cows capable of extended CInt, however, is that the most suitable cows will have the longest CInt, and therefore the fewest number of calves. Farmers in the current study did take this into account when deciding on selection strategy, keeping calves from cows that were persistent in earlier lactations and inseminating less persistent cows with beef bulls (e.g., Belgian Blue) to sell the crossbred calves to the veal industry. Third, farmers aimed for a reduction in farm labor, mainly because of less transition management (i.e., drying-off, calving, start of lactation) and less calf care. Possible positive effects of extended CInt on health, fertility, and farm labor should be subject of further studies to conclude on the viability of customizing CInt on farms.

5 Conclusions

In this study, increased CFSI was not related to SC or conception rate at first AI on 13 commercial Dutch dairy farms that customize calving intervals by increasing the VWP for (part of) their herd. Longer CInt was related to increased SC and decreased conception rate at first AI. On most farms, persistency was greatest in the lowest CInt class (<364 d), probably related to the low peak yield in this class. Excluding this short CInt class, persistency increased with extending CInt on most farms. Though 305-d yield was greater in the longest CInt class (≥532 d) at the majority of farms, effective lactation yield at most farms was greatest in CInt from 364 to 531 d, especially for multiparous cows. Based on the results of this study, it differs per farm what strategy in terms of waiting period for first insemination is optimal for milk production. For heifers on most farms, a CFSI of more than 196 d resulted in greatest effective lactation yield, when high-yielding heifers (differs per farm; >7,500–11,000 kg of FPCM/305 d) were selected. For cows on most farms, a CFSI of more than 140 d resulted in greatest effective lactation yield, when high-yielding cows (differs per farm; >9,500–12,000 kg of FPCM/305 d) were selected.

6 Acknowledgements

The authors thank the participating farmers and CRV (Arnhem, the Netherlands) for providing the data. This study was financed by DairyNL (ZuivelNL; organization of the Dutch dairy supply chain, The Hague, the Netherlands) and the Dutch Ministry of Agriculture, Nature and Food Quality (LNV, The Hague, the Netherlands) as part of the research program ‘One Health for Food’ (1H4F, The Hague, the Netherlands). The authors have not stated any conflicts of interest.

Appendix

Table A1. Number of complete lactations per farm, calving interval (CInt) class[1], and parity class (total lactations = 4,858)

	Parity 1					Parity 2+					
Farm	CInt-1	CInt-2	CInt-3	CInt-4	CInt-5	CInt-1	CInt-2	CInt-3	CInt-4	CInt-5	Total
A	7	47	42	16	14	13	107	62	26	11	345
B	52	62	29	7	5	66	146	64	17	5	453
C	11	23	16	9	19	20	53	40	29	20	240
D	25	39	13	5	7	32	46	25	14	10	216
E	14	41	17	14	11	33	60	53	32	37	312
F	1	48	28	12	2	0	49	58	37	23	258
G	105	141	60	24	19	101	224	126	50	25	875
H	5	157	48	17	13	9	209	104	39	26	627
I	4	15	27	47	70	14	52	72	65	60	426
J	51	51	26	8	12	59	88	44	19	32	390
K	26	40	20	18	9	40	90	56	32	34	365
L	2	21	25	8	21	0	12	31	30	46	196
M	18	22	8	6	4	29	30	20	13	5	155
Total	321	707	359	191	206	416	1,166	755	403	334	4,858

[1] CInt-1 <364; 364 ≤ CInt-2 <420; 420 ≤ CInt-3 <476; 476 ≤ CInt-4 <532; CInt-5 ≥532 d.

Table A2. Mean calving interval (CInt) per farm, CInt class[1], and parity class

	Parity 1					Parity 2+				
Farm	CInt-1	CInt-2	CInt-3	CInt-4	CInt-5	CInt-1	CInt-2	CInt-3	CInt-4	CInt-5
A	348	395	446	505	590	352	390	444	499	570
B	348	390	445	500	544	347	391	446	497	546
C	340	395	446	502	618	347	395	445	500	600
D	349	386	441	506	598	346	385	444	498	570
E	346	396	440	507	639	345	393	445	502	626
F	357	392	445	496	573	NA[2]	399	445	498	577
G	346	386	444	501	583	345	392	444	498	578
H	355	392	441	495	560	346	393	442	497	586
I	342	395	444	506	607	340	397	447	502	592
J	347	384	436	514	587	346	387	440	497	593
K	347	386	446	504	602	346	392	447	503	617
L	341	393	445	493	580	NA	398	443	502	610
M	345	380	435	495	634	339	387	440	497	591

[1] CInt-1 <364; 364 ≤ CInt-2 <420; 420 ≤ CInt-3 <476; 476 ≤ CInt-4 <532; CInt-5 ≥532 d.

[2] NA = not available; in this farm, parity class and CInt class were no lactations.

Table A3. Number of complete lactations per farm, calving to first service interval (CFSI) class[1], and parity class (total lactations = 3,597)

	Parity 1					Parity 2+					
Farm	CFSI-1	CFSI-2	CFSI-3	CFSI-4	CFSI-5	CFSI-1	CFSI-2	CFSI-3	CFSI-4	CFSI-5	Total
C	19	28	18	4	1	29	72	29	8	2	210
D	30	47	5	0	0	66	107	164	212	0	193
E	88	42	19	5	4	50	65	41	22	9	275
F	1	52	22	4	0	1	50	67	26	4	227
G	161	146	12	1	0	170	263	33	2	0	788
H	4	189	14	0	0	11	297	37	4	0	556
I	3	19	38	33	45	16	47	86	49	35	371
J	86	36	9	0	2	111	89	20	3	2	358
K	35	40	11	3	4	73	91	33	10	7	307
L	2	33	25	5	5	1	21	36	25	20	173
M	27	19	4	2	0	46	31	5	3	2	139
Total	386	651	177	57	61	553	1,085	392	154	81	3,597

[1] CFSI-1 <84; 84 ≤ CFSI-2 <140; 140 ≤ CFSI-3 <196; 196 ≤ CFSI-4 <252; CFSI-5 ≥252 d.

Table A4. Mean calving to first service interval (CFSI) per farm, CFSI class[1], and parity class

	Parity 1					Parity 2+				
Farm	CFSI-1	CFSI-2	CFSI-3	CFSI-4	CFSI-5	CFSI-1	CFSI-2	CFSI-3	CFSI-4	CFSI-5
C	61	113	157	226	260	66	115	162	223	267
D	70	103	154	NA[2]	NA	66	107	164	212	NA
E	64	113	157	212	302	62	112	160	218	301
F	79	109	161	260	NA	83	117	162	216	279
G	66	105	157	199	NA	66	107	155	214	NA
H	74	111	157	NA	NA	58	111	155	216	NA
I	57	119	166	222	306	56	115	164	219	292
J	67	104	153	NA	283	66	106	167	238	283
K	70	103	153	236	356	69	109	160	224	290
L	57	111	166	223	294	75	122	168	223	307
M	61	98	166	215	NA	55	101	159	230	303

[1] CFSI-1 <84; 84 ≤ CFSI-2 <140; 140 ≤ CFSI-3 <196; 196 ≤ CFSI-4 <252; CFSI-5 ≥252 d.

[2] NA = not available; in this farm, parity class and CFSI class were no lactations.

Table A5. Significant effects and interaction effects on fat- and protein-corrected milk of the modeled curves on time = t in kilograms per day

Effect[1]	*F*-value	*P*-value
CInt class	22.36	<0.0001
Parity class	3,257.47	<0.0001
Farm	61.48	<0.0001
CInt class × farm	3.76	<0.0001
Parity class × farm	8.52	<0.0001
CInt class × parity class	4.63	0.001
b	4,302.88	<0.0001
b × CInt class	7.23	<0.0001
b × parity class	1,826.08	<0.0001
b × farm	18.37	<0.0001
b × CInt class × farm	2.23	<0.0001
b × parity class × farm	9.17	<0.0001
c	1,007.8	<0.0001
c × parity class	52.46	<0.0001
c × farm	36.06	<0.0001
b_{gest}	1,392.74	<0.0001
b_{gest} × CInt class	5.67	0.0001
b_{gest} × parity class	62.52	<0.0001
b_{gest} × farm	21.93	<0.0001
b_{gest} × CInt class × farm	4.24	<0.0001
b_{gest} × parity class × farm	10.7	<0.0001
CFSI class	27.94	<0.0001
Parity class	1,764.42	<0.0001
Farm	64.87	<0.0001
CFSI class × farm	3.06	<0.0001
Parity class × farm	8.01	<0.0001
CFSI class × parity class	3.62	0.0059
b	8,743.25	<0.0001
b × parity class	1,446.6	<0.0001
b × farm	34.46	<0.0001
b × parity class × farm	8.96	<0.0001
c	347.95	<0.0001
c × CFSI class	2.52	0.0393
c × parity class	13.33	0.0003
c × farm	26.03	<0.0001
c × CFSI class × parity class	3.43	0.0083
b_{gest}	220.91	<0.0001
b_{gest} × CFSI class	2.59	0.035
b_{gest} × parity class	33.48	<0.0001
b_{gest} × farm	8	<0.0001
b_{gest} × CFSI class × farm	5.38	<0.0001
b_{gest} × parity class × farm	12.33	<0.0001

[1] CInt = calving interval; CFSI = calving to first service interval; b = inversely related to persistency; c = related to the beginning of lactation; bgest = inversely related to persistency after gestation effect.

Supplemental tables

Table S1. Peak yield in kg fat- and protein-corrected milk per day, per farm, calving to first service interval (CFSI) class[1], and parity. The shades of green indicate differences between values within one row, where darker green means a higher value

	Parity 1					Parity 2+				
Farm	CFSI-1	CFSI-2	CFSI-3	CFSI-4	CFSI-5	CFSI-1	CFSI-2	CFSI-3	CFSI-4	CFSI-5
C	27.4	27.4	27.5	27.4	30.2	37.0	38.3	39.1	38.9	42.0
D	27.7	28.9	29.8	NA[2]	NA	37.1	39.6	41.2	42.0	NA
E	34.3	36.5	36.1	38.9	38.5	43.1	46.6	46.8	49.3	49.3
F	30.5	31.8	34.9	38.0	NA	42.3	44.9	48.9	51.7	50.0
G	31.7	32.5	32.4	36.1	NA	41.4	43.6	44.2	47.4	NA
H	32.9	33.1	33.8	NA	NA	44.2	45.6	47.0	49.0	NA
I	22.5	28.0	30.1	32.7	32.9	28.8	35.7	38.7	40.9	41.2
J	33.0	33.6	33.3	-[3]	35.2	41.4	43.2	43.5	44.7	45.6
K	29.4	30.7	31.7	35.4	34.0	41.1	44.1	47.2	52.5	47.9
L	19.6	25.1	26.7	-	-	-	-	-	-	-
M	30.3	30.4	31.0	33.5	NA	39.4	41.4	-	-	43.5

[1] CFSI-1 <84; 84 ≤ CFSI-2 <140; 140 ≤ CFSI-3 <196; 196 ≤ CFSI-4 <252; CFSI-5 ≥252 d.

[2] NA: not available; in this farm, parity class, and CFSI class were no lactations.

[3] -: not possible to compute this value with the lactation curves.

Table S2. Persistency (decrease in kg fat- and protein-corrected milk per day) per farm, calving to first service interval (CFSI) class[1], and parity. The shades of green indicate differences between values within one row, where darker green means a higher value

	Parity 1					Parity 2+				
Farm	CFSI-1	CFSI-2	CFSI-3	CFSI-4	CFSI-5	CFSI-1	CFSI-2	CFSI-3	CFSI-4	CFSI-5
C	-0.0072	-0.0072	-0.0071	-0.0075	-0.0074	-0.0566	-0.0566	-0.0567	-0.0568	-0.0566
D	-0.0438	-0.0438	-0.0437	NA[2]	NA	-0.0735	-0.0736	-0.0736	-0.0737	NA
E	-0.0294	-0.0293	-0.0293	-0.0297	-0.0296	-0.0586	-0.0587	-0.0588	-0.0588	-0.0587
F	-0.0181	-0.0180	-0.0179	-0.0183	NA	-0.0664	-0.0664	-0.0665	-0.0666	-0.0664
G	-0.0306	-0.0306	-0.0305	-0.0309	NA	-0.0686	-0.0686	-0.0687	-0.0688	NA
H	-0.0304	-0.0303	-0.0303	NA	NA	-0.0679	-0.0680	-0.0681	-0.0681	NA
I	-0.0267	-0.0266	-0.0266	-0.0269	-0.0269	-0.0554	-0.0554	-0.0555	-0.0556	-0.0554
J	-0.0309	-0.0308	-0.0308	NA	-0.0311	-0.0618	-0.0619	-0.0620	-0.0620	-0.0619
K	-0.0203	-0.0202	-0.0201	-0.0205	-0.0204	-0.0603	-0.0603	-0.0604	-0.0604	-0.0603
L	-0.0147	-0.0147	-0.0146	-0.0150	-0.0149	-0.0377	-0.0378	-0.0378	-0.0379	-0.0378
M	-0.0240	-0.0239	-0.0239	-0.0243	NA	-0.0495	-0.0496	-0.0497	-0.0497	-0.0496

[1] CFSI-1 <84; 84 ≤ CFSI-2 <140; 140 ≤ CFSI-3 <196; 196 ≤ CFSI-4 <252; CFSI-5 ≥252 d.

[2] NA: not available; in this farm, parity class, and CFSI class were no lactations.

Table S3. 305-d yield in kg fat- and protein-corrected milk per farm, calving to first service interval (CFSI) class[1], and parity. The shades of green indicate differences between values within one row, where darker green means a higher value

	Parity 1					Parity 2+				
Farm	CFSI-1	CFSI-2	CFSI-3	CFSI-4	CFSI-5	CFSI-1	CFSI-2	CFSI-3	CFSI-4	CFSI-5
C	8,019	7,994	8,012	8,067	8,902	9,488	9,844	10,036	9,936	10,954
D	7,121	7,462	7,725	NA[2]	NA	9,048	9,760	10,196	10,422	NA
E	9,598	10,181	10,031	10,933	10,805	11,440	12,375	12,400	13,147	13,202
F	8,827	9,105	10,026	11,010	NA	10,698	11,458	12,557	13,386	13,005
G	8,774	8,950	8,903	10,034	NA	10,493	11,069	11,196	12,172	NA
H	9,091	9,110	9,320	NA	NA	11,486	11,771	12,160	12,758	NA
I	6,090	7,715	8,348	9,096	9,176	7,004	8,990	9,796	10,388	10,652
J	9,122	9,229	9,137	NA	9,755	10,739	11,245	11,327	11,683	11,972
K	8,339	8,723	9,032	10,051	9,676	10,379	11,129	11,618	12,482	12,290
L	5,512	7,193	7,686	7,873	7,987	7,670	9,715	10,382	10,414	10,711
M	8,536	8,527	8,710	9,169	NA	10,304	10,622	10,979	11,282	11,252

[1] CFSI-1 <84; 84 ≤ CFSI-2 <140; 140 ≤ CFSI-3 <196; 196 ≤ CFSI-4 <252; CFSI-5 ≥252 d.

[2] NA: not available; in this farm, parity class, and CFSI class were no lactations.

Table S4. Effective lactation yield in kg fat- and protein-corrected milk per day of calving interval per farm, calving to first service interval (CFSI) class[1], and parity. The shades of green indicate differences between values within one row, where darker green means a higher value

	Parity 1					Parity 2+				
Farm	CFSI-1	CFSI-2	CFSI-3	CFSI-4	CFSI-5	CFSI-1	CFSI-2	CFSI-3	CFSI-4	CFSI-5
C	23.9	24.2	24.4	23.6	27.7	26.7	27.1	26.7	24.6	28.5
D	20.3	20.9	20.6	NA[2]	NA	25.4	26.8	26.1	24.6	NA
E	28.0	29.9	28.9	30.8	29.1	33.1	35.2	34.2	34.8	33.3
F	25.6	26.7	29.6	32.5	NA	30.1	32.2	34.2	35.8	33.4
G	25.5	26.1	25.4	29.0	NA	29.9	31.1	30.0	33.2	NA
H	26.5	26.7	26.8	NA	NA	33.2	33.5	33.2	33.7	NA
I	17.6	22.1	23.8	25.6	25.1	20.1	25.1	26.5	27.5	26.6
J	26.6	26.9	26.2	NA	27.6	30.8	31.4	30.6	30.1	30.9
K	24.8	25.9	26.7	29.4	27.0	29.7	31.4	31.9	32.9	30.1
L	16.1	21.2	22.3	22.4	22.8	22.5	28.3	29.2	28.6	28.3
M	25.0	24.9	25.2	26.4	NA	29.9	30.4	30.8	30.6	28.4

[1] CFSI-1 <84; 84 ≤ CFSI-2 <140; 140 ≤ CFSI-3 <196; 196 ≤ CFSI-4 <252; CFSI-5 ≥252 d.

[2] NA: not available; in this farm, parity class, and CFSI class were no lactations.

Table S5. Peak yield in kg fat- and protein-corrected milk per day, per farm, calving interval (CInt) class[1], and parity. The shades of green indicate differences between values within one row, where darker green means a higher value

	Parity 1					Parity 2+				
Farm	CInt-1	CInt-2	CInt-3	CInt-4	CInt-5	CInt-1	CInt-2	CInt-3	CInt-4	CInt-5
A	26.9	28.5	29.4	29.7	29.3	36.8	39.4	40.3	41.0	41.1
B	29.9	31.8	34.1	34.1	31.5	41.1	43.9	46.3	46.8	44.6
C	27.1	27.6	27.0	27.4	28.7	37.3	38.3	37.5	38.4	40.7
D	27.2	28.8	30.2	27.7	29.5	36.7	39.0	40.6	38.5	40.8
E	32.5	36.3	37.6	37.5	37.6	40.8	45.6	47.1	47.6	48.2
F	28.9	31.2	34.3	35.8	37.1	NA[2]	44.2	47.9	49.7	51.4
G	31.4	32.8	33.1	31.9	32.4	41.0	43.1	43.5	43.4	43.8
H	30.2	33.3	33.4	33.6	33.1	41.6	45.5	45.9	46.4	46.4
I	20.3	27.5	30.7	32.4	31.8	26.8	35.2	38.4	40.7	40.6
J	32.8	33.1	34.9	34.6	31.3	41.3	42.3	44.3	44.4	41.6
K	28.3	30.2	31.0	31.7	32.2	40.1	43.1	43.7	44.9	46.0
L	-[3]	25.2	26.1	26.9	27.0	NA	-	-	-	-
M	30.3	30.5	29.8	31.7	31.8	39.7	40.4	39.9	42.5	42.9

[1] CInt-1 <364; 364 ≤ CInt-2 <420; 420 ≤ CInt-3 <476; 476 ≤ CInt-4 <532; CInt-5 ≥532 d.

[2] NA: not available; in this farm, parity class, and CInt class were no lactations.

[3] -: not possible to compute this value with the lactation curves.

Table S6. Persistency (decrease in kg fat- and protein-corrected milk per day) per farm, calving interval (CInt) class[1], and parity. The shades of green indicate differences between values within one row, where darker green means a higher value

	Parity 1					Parity 2+				
Farm	CInt-1	CInt-2	CInt-3	CInt-4	CInt-5	CInt-1	CInt-2	CInt-3	CInt-4	CInt-5
A	-0.018	-0.024	-0.022	-0.020	-0.016	-0.046	-0.052	-0.051	-0.049	-0.045
B	-0.018	-0.026	-0.026	-0.025	-0.018	-0.051	-0.059	-0.059	-0.058	-0.052
C	-0.014	-0.008	-0.003	-0.003	-0.008	-0.065	-0.059	-0.055	-0.054	-0.059
D	-0.048	-0.043	-0.050	-0.039	-0.030	-0.077	-0.073	-0.079	-0.069	-0.059
E	-0.012	-0.029	-0.034	-0.032	-0.029	-0.042	-0.059	-0.064	-0.061	-0.058
F	-0.052	-0.013	-0.026	-0.020	-0.013	NA[2]	-0.063	-0.075	-0.070	-0.063
G	-0.030	-0.034	-0.030	-0.026	-0.023	-0.068	-0.072	-0.068	-0.064	-0.061
H	-0.023	-0.030	-0.032	-0.029	-0.022	-0.062	-0.069	-0.071	-0.067	-0.060
I	-0.013	-0.028	-0.028	-0.031	-0.025	-0.042	-0.057	-0.057	-0.060	-0.053
J	-0.033	-0.032	-0.037	-0.033	-0.022	-0.065	-0.063	-0.068	-0.064	-0.053
K	-0.016	-0.026	-0.020	-0.019	-0.017	-0.056	-0.065	-0.059	-0.059	-0.056
L	-[3]	-0.015	-0.013	-0.012	-0.015	NA	-0.040	-0.038	-0.036	-0.040
M	-0.030	-0.021	-0.024	-0.029	-0.018	-0.054	-0.045	-0.049	-0.053	-0.043

[1] CInt-1 <364; 364 ≤ CInt-2 <420; 420 ≤ CInt-3 <476; 476 ≤ CInt-4 <532; CInt-5 ≥532 d.

[2] NA: not available; in this farm, parity class, and CInt class were no lactations.

[3] -: not possible to compute this value with the lactation curves.

Table S7. 305-d yield in kg fat- and protein-corrected milk per farm, calving interval (CInt) class[1], and parity. The shades of green indicate differences between values within one row, where darker green means a higher value

	Parity 1					Parity 2+				
Farm	CInt-1	CInt-2	CInt-3	CInt-4	CInt-5	CInt-1	CInt-2	CInt-3	CInt-4	CInt-5
A	7,672	7,948	8,250	8,399	8,425	9,486	9,888	10,192	10,503	10,722
B	8,737	8,939	9,583	9,618	9,027	11,110	11,352	12,008	12,205	11,808
C	7,982	8,097	7,957	8,073	8,379	9,292	9,771	9,683	9,960	10,461
D	6,800	7,462	7,677	7,222	8,038	8,820	9,644	9,872	9,578	10,589
E	9,705	10,145	10,362	10,417	10,525	11,547	12,130	12,359	12,575	12,878
F	7,001	9,060	9,664	10,272	10,843	NA[2]	11,370	12,011	12,781	13,546
G	8,848	8,983	9,156	8,889	9,121	10,577	10,872	11,085	11,173	11,405
H	8,566	9,217	9,168	9,303	9,330	11,107	11,789	11,744	12,040	12,262
I	5,822	7,517	8,476	8,899	8,915	6,867	8,712	9,697	10,282	10,492
J	9,156	9,106	9,478	9,478	8,770	10,851	10,987	11,395	11,556	11,043
K	8,464	8,479	8,838	9,067	9,290	10,489	10,675	11,085	11,474	11,892
L	6,384	7,177	7,483	7,800	7,689	NA	9,672	9,994	10,473	10,556
M	8,483	8,742	8,334	8,744	9,132	10,347	10,662	10,293	10,865	11,447

[1] CInt-1 <364; 364 ≤ CInt-2 <420; 420 ≤ CInt-3 <476; 476 ≤ CInt-4 <532; CInt-5 ≥532 d.

[2] NA: not available; in this farm, parity class, and CInt class were no lactations.

Table S8. Effective lactation yield in kg fat- and protein-corrected milk per day of calving interval (CInt), per farm, CInt class[1], and parity. The shades of green indicate differences between values within one row, where darker green means a higher value

	Parity 1					Parity 2+				
Farm	CInt-1	CInt-2	CInt-3	CInt-4	CInt-5	CInt-1	CInt-2	CInt-3	CInt-4	CInt-5
A	22.1	23.0	23.8	24.2	24.3	27.3	28.4	28.6	28.7	28.6
B	25.2	26.1	27.8	27.9	26.2	32.0	32.7	33.7	33.6	31.6
C	22.9	24.1	24.2	24.9	25.0	26.8	27.6	26.7	26.6	25.0
D	19.6	21.3	20.7	19.0	21.4	25.4	27.2	26.0	24.2	26.5
E	27.9	29.8	29.8	29.9	28.9	33.2	35.1	34.6	34.4	32.5
F	19.9	26.6	28.1	30.1	32.6	NA[2]	32.3	32.8	34.2	35.5
G	25.5	26.1	26.5	25.6	26.0	30.5	31.0	30.6	29.9	29.0
H	24.8	27.0	26.5	26.6	26.8	32.0	34.1	32.7	32.4	31.5
I	16.7	21.6	24.3	24.9	24.7	19.8	24.6	26.7	27.1	26.4
J	26.4	26.5	27.3	26.6	25.1	31.3	31.5	31.7	30.9	28.4
K	24.4	24.9	26.3	26.8	26.9	30.2	30.5	31.1	31.2	29.7
L	18.2	21.1	21.7	22.9	22.0	NA	28.3	28.5	29.6	27.9
M	24.4	25.7	24.0	24.6	26.1	29.8	31.1	29.0	29.4	30.7

[1] CInt-1 <364; 364 ≤ CInt-2 <420; 420 ≤ CInt-3 <476; 476 ≤ CInt-4 <532; CInt-5 ≥532 d.

[2] NA: not available; in this farm, parity class, and CInt class were no lactations.

Chapter 3

Effects of extended voluntary waiting period from calving until first insemination on body condition, milk yield, and lactation persistency

E.E.A. Burgers[1,2], A. Kok[1,3], R.M.A. Goselink[2], H. Hogeveen[4], B. Kemp[1], A.T.M. van Knegsel[1]

[1]Adaptation Physiology Group, Wageningen University & Research, Wageningen, the Netherlands
[2]Wageningen Livestock Research, Wageningen University & Research, Wageningen, the Netherlands
[3]Animal Production Systems Group, Wageningen University & Research, Wageningen, the Netherlands
[4]Business Economics Group, Wageningen University & Research, Wageningen, the Netherlands

Journal of Dairy Science 104 (2021): 8009 – 8022

Abstract

A 1-yr calving interval (CInt) is usually associated with maximized milk output, due to the calving-related peak in milk yield. Extending CInt could benefit cow health and production efficiency due to fewer transition periods per unit of time. Extending CInt can affect lactation performance by fewer days dry per year, delayed pregnancy effect on milk yield, and greater milk solid yield in late lactation. This study first investigated the effects of 3 different voluntary waiting periods (VWP) from calving until first insemination on body weight, body condition, milk yield, and lactation persistency. Second, individual cow characteristics in early lactation were identified that contributed to milk yield and persistency of cows with different VWP. Holstein-Friesian dairy cows (n = 154) within 1 herd were blocked for parity, calving season, and expected milk yield. Cows were randomly assigned within the blocks to 1 of 3 VWP (50, 125, or 200 d: VWP50, VWP125, or VWP200, respectively) and monitored through 1 complete lactation and the first 6 wk of the subsequent lactation, or until culling. Minimum and mean CInt (384 vs. 452 vs. 501 d for VWP50 vs. VWP125 vs. VWP200) increased with increasing VWP, but maximum CInt was equal for the 3 VWP. Fat- and protein-corrected milk yield (FPCM) was analyzed weekly. Milk yield and FPCM were also expressed per day of CInt, to compare yields of cows with different VWP. Persistency was determined between d 100 and d 200 of the lactation, as well as between d 100 and dry-off. Values are presented as least squares means ± standard error of the mean. During the first 44 wk of lactation, VWP did not affect FPCM yield in both primiparous and multiparous cows. The VWP did not affect milk yield per day of CInt. The VWP did not affect FPCM yield per day of calving interval for primiparous cows. Multiparous cows in VWP125 had FPCM yield per day of CInt similar to that of VWP50. Multiparous cows in VWP200 had lower FPCM yield per day of CInt compared with VWP50 (27.2 vs. 30.4 kg/d). During the last 6 wk before dry-off, cows in VWP125 had lower yield compared with cows in VWP50, which could benefit their udder health in the dry period and after calving. Persistency was better for cows in VWP200 compared with cows in VWP50 (−0.05 vs. −0.07 kg/d). Body weight was not different among VWP groups. Multiparous cows in VWP200 had a higher body condition score in the last 3 mo before dry-off and the first 6 wk of the next lactation, compared with multiparous cows in VWP125 and VWP50. The VWP could be extended from 50 d to 125 d without an effect on daily yield per day of calving interval. Extending VWP until 200 d for primiparous cows did not affect their daily milk yield, but multiparous cows with a 200-d VWP had a reduced milk yield per day of calving interval and an increased body condition in late lactation and the subsequent lactation, compared with multiparous cows with a 50-d VWP.

Key words: extended calving interval, milk production, lactation persistency, individual cow variation

1 Introduction

Traditionally, farmers aim for a 1-yr calving interval (CInt) for their cows, as calving is associated with a peak in milk yield around wk 4 to 7 of lactation (Butler et al., 1981). Around calving, however, cows experience multiple transitions, such as drying-off, calving itself, and the start of the next lactation. During these transitions, cows have an increased risk for developing diseases and disorders, such as mastitis, hypocalcemia, and ketosis (Friggens et al., 2004). With a 1-yr CInt, cows face these transitions every year. Moreover, high-yielding cows in a 1-yr CInt often have milk yields above 18 kg at the moment of dry-off, which increases the risk for udder infections in the dry period and at calving (Odensten et al., 2007).

To reduce the number of transitions per unit of time, CInt can be extended by extending the voluntary waiting period (VWP) from calving until first insemination. Moreover, extending VWP from 40 to 180 d resulted in a greater proportion (34.2 vs. 54.6%) of cows being dried off at lower milk yields (<15 kg; Niozas et al., 2019a), which could be beneficial for udder health (Rajala-Schultz et al., 2005; Odensten et al., 2007) and cow welfare (Zobel et al., 2015). Cows with an extended CInt, however, have fewer peaks in milk yield per unit of time compared with cows with a 1-yr CInt. This could result in a lower milk yield per cow per year. Most studies that analyzed farm data retrospectively found that cows in longer CInt had a lower yearly milk yield compared with cows in shorter CInt (Strandberg and Oltenacu, 1989; Inchaisri et al., 2010; Kok et al., 2019). In such retrospective analyses of farm data, however, extended CInt may be the consequence of health and fertility problems, and cows are not deliberately managed for an extended CInt (Mellado et al., 2016). Moreover, analyses often focused on the 305-d milk yield (Strandberg and Oltenacu, 1989; Steeneveld and Hogeveen, 2012). Cows in an extended CInt, however, have longer lactation periods and fewer days dry per year, which both influence the average milk yield per day and per year (Kok et al., 2019). As an alternative to 305-d milk yield, milk yield could be expressed as the milk yield per day of CInt (Kok et al., 2016; Lehmann et al., 2016), which would account for longer lactation periods or differences in days dry per year.

The CInt has been deliberately extended in experimental studies (Rehn et al., 2000; Arbel et al., 2001; Niozas et al., 2019a) and on commercial farms (Lehmann et al., 2016). When CInt was extended from 368 to 430 d for both primiparous and multiparous cows, milk yield and ECM per day of CInt did not differ (Rehn et al., 2000). Niozas et al. (2019a) also reported a similar milk yield and ECM per day of CInt for cows with VWP of 40, 120, and 180 d, and an increase in lactation persistency for the cows with an extended lactation. Milk yield before dry-off was reduced for cows with a VWP of 180 d compared with a VWP of 40 or 120 d (Niozas et al., 2019a). At the time of dry-off, however, cows with a VWP of 180 d had a higher BCS compared with cows with a VWP of 40 or 120 d, which could negatively affect their health in the subsequent lactation (Roche and Berry, 2006). Fat and protein content were similar (Rehn et al., 2000) or greater (Österman and Bertilsson, 2003) in longer CInt compared with shorter CInt. This could be attributed to an increase in fat and protein content later in lactation (Silvestre et al., 2009). An increase in fat and protein content in longer CInt might compensate for possible lower milk yield.

Parity could affect the results for milk yield of cows after different VWP. When extending CInt for primiparous cows, milk and ECM yield per day of CInt increased, whereas for multiparous cows, yield stayed the same (Arbel et al., 2001; Lehmann et al., 2016) or decreased (Österman and Bertilsson, 2003). Differences among parities concerning consequences of an extended VWP on milk yield could be related to more persistent lactations for primiparous cows compared with multiparous cows (Niozas et al., 2019a). Besides parity, other characteristics of individual cows, such as peak yield or persistency, can also be hypothesized to affect milk yield of cows after different VWP. Knowledge on relevant individual cow characteristics related to milk yield after different VWP could support selection of cows for different VWP.

The first aim of this study was to investigate the effect of 3 VWP (50, 125, and 200 d) on BW, body condition, milk and solids yield, and lactation persistency. The second aim was to see how individual cow characteristics in early lactation, such as parity, maximum yield, time of maximum yield, and slope to maximum yield affect milk yield per day of CInt and persistency in cows with different VWP.

2 Materials and methods

2.1 Animals and housing

The experimental protocol was approved by the Institutional Animal Care and Use Committee of Wageningen University & Research (the Netherlands) and complies with the Dutch law on Animal Experimentation (protocol number 2016.D-0038.005). The experiment was conducted at Dairy Campus research farm (Leeuwarden, the Netherlands) between December 2017 and January 2020.

Cows were selected from a research herd of 500 lactating Holstein-Friesian cows based on the following criteria: no twin pregnancy, no clinical mastitis or SCC >250,000 at the final 2 milk test days before dry-off and expected to finish a complete lactation based on being in good general health. The experimental period started at calving and ended 6 wk after the next calving, or at 530 DIM if cows were not pregnant. Cows that were culled were followed until they were culled. Cows were milked twice daily around 0600 h and 1800 h in a 40-cow rotary milking parlor (GEA). Partial mixed ration during lactation consisted of grass silage, corn silage, soybean meal, and wheat meal, supporting 22 kg of milk. Concentrate supply started at 1 kg/d on the day of calving and increased stepwise to 9 kg/d for primiparous cows or 10 kg/d for multiparous cows from d 21 onward. After 100 DIM, individual concentrate supply was decreased to match reductions in milk yield based on the last 5 d of milk yield. In the milking parlor, 1 kg of additional concentrate was supplied daily. Ration during the dry period consisted of grass silage and corn silage, supplemented with wheat straw and concentrate. In the last 10 d before the expected calving date, cows received 1 kg of concentrate daily. Cows were dried off between 42 and 49 d before the expected calving date. From 7 d before dry-off, cows were given the dry-cow ration. The 3 d before dry-off, cows were milked once daily. When cows had SCC >150,000 at the final milk test day, cows were treated with antibiotics at dry-off (Orbenin Dry Cow Extra, Zoetis). All cows were treated with teat sealant at dry-off (Orbeseal, Zoetis).

2.2 Experimental design

In total, 154 cows were selected (41 primiparous, 113 multiparous). In wk 6 after calving, cows were blocked for parity, calving date, 305-d milk yield in the previous lactation (multiparous cows) or expected milk yield (primiparous cows), and breeding value for persistency (CRV). Each block consisted of 3 cows. First, 50 blocks of 3 cows were formed. After removal of 2 cows before the end of VWP due to culling as a result of diseases, 2 more blocks of 3 cows were added. Mean ranges within blocks of the variables used to block the cows are presented in Appendix Table A1. The cows were divided randomly within blocks over 3 treatment groups: a VWP of 50 d (VWP50), 125 d (VWP125), or 200 d (VWP200), resulting in equal absolute difference in days between the 3 groups. Cows in the 3 treatment groups were inseminated after their VWP when estrus was detected. Estrus detection was carried out by using the Nedap

Smarttag system as well as visually by the animal caretaker. Cows were inseminated until 300 DIM, meaning that cows in VWP50 had 250 d to conceive, cows in VWP125 had 175 d to conceive, and cows in VWP200 had 100 d to conceive. Cows that did not conceive within 300 DIM stayed in the experiment until 530 DIM as long as they produced at least 10 L of milk per day. Cows left the experiment when milk yield dropped below 10 L of milk per day based on evaluation of daily milk yield in the preceding 7 d. As a consequence of this approach, 6 cows that did not conceive left the experiment before 530 DIM.

2.3 Measurements and calculations

Milk yield was recorded at every milking from day of calving until dry-off and the first 6 wk of the next lactation. Milk samples for the analysis of fat, protein, and lactose were collected for each individual cow from the container 4 times per week (Tuesday p.m., Wednesday a.m., Wednesday p.m., Thursday a.m.) in 10-mL tubes containing Bronopol as a preservative and analyzed for the percentage of fat, protein, and lactose as a pooled sample (ISO, 2013; Qlip, Zutphen, the Netherlands). Body condition score was visually evaluated every 4 wk by the same person using a 1 to 5 scale (Ferguson et al., 1994). Body weight was recorded twice daily after milking, by a scale that the cows walked over when returning from the milking rotary to the pen (GEA).

Milk production was converted to fat- and protein-corrected milk (FPCM) using the following formula (CVB, 2012):

$$\mathrm{FPCM(kg)} = \mathrm{milk\ (kg)} \times (0.337 + 0.116 \times \mathrm{fat\ (\%)} + 0.06 \times \mathrm{protein\ (\%)}),$$

using the weekly contents of fat and protein, and the mean daily milk yield of each week. Milk yield and FPCM yield per day of CInt were calculated for each individual cow for the entire CInt, or from calving until culling. Mean fat, protein, and lactose content was calculated by summing the contents for the entire first lactation within the experiment and dividing them by the number of measurements. Fat, protein, and lactose yields were, similar to milk and FPCM, calculated as kilograms of yield per day of CInt in the first lactation within the experiment. The individual milk or FPCM yield relative to mean milk or FPCM yield in the first 6 wk of lactation of all primiparous or multiparous cows in the experiment (relative yield) was calculated, to include as a covariate in the statistical analysis in some of the models. The first 6 wk are the period before VWP treatment started, so if production differed between groups during this time, their production in the rest of lactation could be corrected with the relative yield in early lactation.

Lactation persistency was defined as the reduction in milk yield after peak yield and was calculated over 2 different intervals as the slope between (1) d 100 to 200 in lactation and (2) d 100 to start of dry-off ration (7 d before the dry-off date; Chen et al., 2016). Day 100 to 200 was chosen as the period for a standardized persistency for each cow, because no effect of pregnancy on the lactation curve was expected during this period. Day 100 to the start of dry-off was chosen because a difference was expected between short and extended VWP, mainly due to a delayed effect of pregnancy on the lactation curve after extended VWP (Strandberg and Lundberg, 1991). To calculate the slope, first a 2-sided moving average was made of the milk yield between 5 d around d 100 and around d 200 separately (adjusted from Poppe et al., 2020). In this way, to calculate lactation persistency, milk yield at d 100 and d 200 was defined as the mean milk yield of the 2 d before, the 2 d after, and the day itself. The moving average of the milk yield at the start of dry-off was calculated over the 5 d before the start of dry-off. The moving average was used instead of the daily milk yield records to reduce the effects of daily fluctuations in milk yield on persistency measures.

To evaluate individual cow characteristics that predict cow performance after different VWP, several early-lactation curve characteristics were determined for each cow for the first 6 wk of lactation. First, maximum yield in this period was defined per animal as the greatest 5-d rolling average milk yield in the first 6 wk of the first lactation within the experiment. Second, day of maximum yield was defined as the day around which the 5-d rolling average yield was greatest, and divided into 3 classes (≤30 d, 31–35 d, and 36–42 d). Third, slope to maximum yield was defined as the slope per day from d 10 in lactation until day of maximum yield and was computed as maximum yield minus the 5-d rolling average of the milk yield on d 10 in lactation (i.e., the average milk yield from d 8 until d 12), divided by the day of maximum yield minus 10. Slope to maximum yield was log-transformed to meet the requirement of a normal distribution.

Next to these lactation curve characteristics, the mean FPCM yield, the mean milk yield, fat, protein, and lactose content, fat-to-protein ratio, BCS, and BW were determined per cow for the first 6 wk in lactation.

2.4 Statistical analysis

Visual inspection of the data indicated normality; non-normally distributed data were transformed. Parity class (primiparous or multiparous cows) refers to the parity of the cow during the first lactation within the experiment.

Model 1: A general linear mixed model (PROC MIXED, SAS version 9.4, SAS Institute Inc.) was used to test the effects of VWP on the dependent variables: CInt (model 1a), dry period length (model 1b), and interval length (i.e., calving interval or interval from calving until the cow left the experiment; model 1c):

$$y_i = \mu + \mathrm{VWP}_i + \varepsilon_i, \qquad [1]$$

where y_i represents the dependent variables, μ represents the mean, VWP_i represents the VWP (i = 50, 125, or 200 d), and $\boldsymbol{\varepsilon}_i$ represents the random residual term from a normal distribution. Parity class was not included in these models, as preliminary analyses showed it was not significant.

Model 2: A general linear mixed model (PROC MIXED, SAS version 9.4) was used to test the effects of VWP and parity class on the dependent variables: lactation yield per day of CInt [milk (model 2a), FPCM (model 2b), fat (model 2c), protein (model 2d), lactose (model 2e)] and persistency [d 100–200 (model 2e) and d 100 to start of dry-off ration (model 2f)]:

$$y_{ij} = \mu + \mathrm{VWP}_i + \mathrm{Par}_j + (\mathrm{VWP} \times \mathrm{Par})_{ij} + \varepsilon_{ij}, \qquad [2]$$

where y_{ij} represents the dependent variables, μ represents the mean, VWP_i represents the VWP (i = 50, 125, or 200 d), Par_j represents the parity class (j = 1 or 2+), $(\mathrm{VWP} \times \mathrm{Par})_{ij}$ represents the interaction between VWP and parity class, and $\boldsymbol{\varepsilon}_{ij}$ represents the random residual term from a normal distribution.

Models 3 and 4: These models were adjusted from models 2a and 2b but additionally included relative yield as covariate. These models were performed both including (models 3a and 3b) and excluding (models 4a and 4b) cows that did not become pregnant or were culled in the experiment:

$$y_{ijk} = \mu + \mathrm{VWP}_i + \mathrm{Par}_j + (\mathrm{VWP} \times \mathrm{Par})_{ij} + \text{Relative Yield}_k + \varepsilon_{ijk}, \qquad [3,4]$$

where Relative Yield$_k$ represents the individual milk yield relative to mean yield in the first 6 wk of lactation of all primiparous or multiparous cows in the experiment.

Model 5: A Pearson correlation (PROC CORR, SAS version 9.4) was used to test the relation between persistency between d 100 and 200 and persistency between d 100 and the start of dry-off, for each VWP × parity class combination and for each VWP class.

Model 6: A repeated measurements model in SAS (PROC MIXED, SAS version 9.4) was used to test the effects of VWP and parity class on the dependent variables milk yield (model 6a), FPCM (model 6b), fat content (model 6c), protein content (model 6d), lactose content (model 6e), and BW (model 6f):

$$y_{ijk} = \mu + \text{VWP}_i + \text{Par}_j + \text{Week}_k + (\text{VWP} \times \text{Par})_{ij} + (\text{VWP} \times \text{Week})_{ik} + (\text{Par} \times \text{Week})_{jk} + \varepsilon_{ijk}, \qquad [6]$$

where y_{ijk} represents the dependent variable, μ represents the mean, VWP$_i$ represents the VWP (i = 50, 125, or 200 d), Par$_j$ represents the parity class (j = 1 or 2+), Week$_k$ represents the lactation week from the first calving within the experiment (1, 2, 3, …, 6; 1, 2, 3, …, 44) or lactation week relative to the second calving within the experiment (−6, −5, …, −1 or −12, −11, …, −1; 1, 2, 3, …, 6), (VWP × Par)$_{ij}$ represents the interaction between VWP and parity class, (VWP × Week)$_{ik}$ represents the interaction between VWP and lactation week, (Par × Week)$_{jk}$ represents the interaction between parity class and lactation week, and ε_{ijk} represents the random residual term from a normal distribution. The model included a repeated measurement effect of lactation weeks with cow as the repeated subject. The same repeated measurements model was used to test the effects of VWP and parity class on the dependent variable BCS (model 6g):

$$y_{ijk} = \mu + \text{VWP}_i + \text{Par}_j + \text{Month}_k + (\text{VWP} \times \text{Par})_{ij} + (\text{VWP} \times \text{Month})_{ik} + (\text{Par} \times \text{Month})_{jk} + \varepsilon_{ijk}, \qquad [6]$$

where Month$_k$ represents the lactation month from the first calving within the experiment (1 or 2; 1, 2, 3, …, 11) or lactation month relative to the second calving within the experiment (−3, −2, −1; 1 or 2).

Models 7 and 8: These models were adjusted from models 6a and 6b, additionally including relative yield as covariate. These models were performed both including (models 7a and 7b) and excluding (models 8a and 8b) cows that did not become pregnant or were culled in the experiment:

$$y_{ijk} = \mu + \text{VWP}_i + \text{Par}_j + \text{Week}_k + \text{Relative Yield}_l + (\text{VWP} \times \text{Par})_{ij} + (\text{VWP} \times \text{Week})_{ik} + (\text{Par} \times \text{Week})_{jk} + \varepsilon_{ijkl}. \qquad [7, 8]$$

Models 9 and 10: A general linear model was used to predict individual FPCM yield per day of CInt (model 9) and lactation persistency from d 100 until the start of dry-off (model 10) after different VWP. The following cow characteristics in early lactation (first 6 wk) were tested: maximum yield, day of maximum yield, slope to maximum yield, mean FPCM yield, mean milk yield, fat, protein, and lactose content, fat-to-protein ratio, BCS, and BW. Next to these early-lactation characteristics, calving date, expected (primiparous cows) or previous (multiparous cows) 305-d milk yield, and breeding value for persistency were tested. First, the effects of each cow characteristic on FPCM yield per day of CInt and on lactation persistency were tested with a univariate analysis, using a general linear mixed model in SAS (PROC MIXED). Second, when *P*-value was < 0.2, the characteristic was included in the multivariate model. The multivariate model always included VWP and parity class as fixed effects. The cow characteristics in early lactation and their interaction with VWP and parity class stayed in the model if $P < 0.05$ by using backward selection.

Model 11: To evaluate the effect of CInt, 3 equal groups with different CInt length were formed of cows that completed the CInt within the experiment: <415 d (n = 43), 415 to 485 d (n = 44), or >485 d (n = 40). A general linear mixed model (PROC MIXED) was used to test the effects of CInt group and parity class on the dependent variables: lactation yield per day of CInt [milk (model 11a), FPCM (model 11b)] and persistency [d 100 to start of dry-off ration (model 11c)]:

$$y_{ij} = \mu + \text{CInt}_i + \text{Par}_j + (\text{CInt} \times \text{Par})_{ij} + \varepsilon_{ij}, \qquad [11]$$

where y_{ij} represents the dependent variables, μ represents the mean, CInt_i represents the CInt group (i is <415, 415–485, or >485 d), Par_j represents the parity class (j = 1 or 2+), $(\text{CInt} \times \text{Par})_{ij}$ represents the interaction between CInt group and parity class, and $\boldsymbol{\varepsilon}_{ij}$ represents the random residual term from a normal distribution. Results of this model are presented in Appendix Table A2.

Values are presented as least squares means ± standard error of the mean. All *P*-values of pair-wise comparisons of least squares means were corrected with a Bonferroni adjustment.

3 Results

From the 154 cows that entered the experiment, 127 cows started a second lactation within the experiment. These cows were followed for a complete lactation and 6 wk into the next lactation. In total, 14 cows did not become pregnant during the first lactation (2 from VWP50, 3 from VWP125, 9 from VWP200), and 13 cows were culled due to health issues (5 from VWP50, 4 from VWP125, 4 from VWP200). Cows that were culled before the end of the experiment were followed until they were culled. Excluding culled and nonpregnant cows, calving interval was 384 (±6.75), 452 (±7.14), or 501 (±7.50) d for cows in VWP50, VWP125, or VWP200 (Table 1). Dry period length did not differ among the 3 VWP groups. Including culled and nonpregnant cows, interval length was 363 (±12.2), 445 (±12.8), or 481 (±12.5) d for cows in VWP50, VWP125, or VWP200 (Table 2).

Table 1. Calving interval (CInt, d) and dry period length (DP, d) of the 127 cows that had a second calf within the experiment and had a voluntary waiting period after calving until first insemination of 50, 125, or 200 d (VWP50, VWP125, or VWP200)[1]

Waiting period	CInt	Range	SEM	DP	Range	SEM
VWP50	384^{a}	324–565	6.75	41	18–63	1.4
VWP125	452^{b}	400–586	7.14	42	8–72	1.5
VWP200	501^{c}	469–575	7.50	43	8–75	1.6

$^{a-c}$Different superscript letters indicate a difference among LSM within the column (a-b: $P < 0.01$; a-c: $P < 0.01$; b-c: $P < 0.01$).

[1] Values represent LSM, range, and SEM.

Table 2. Interval length (d) of all 154 cows within the experiment that had a voluntary waiting period after calving until first insemination of 50, 125, or 200 d (VWP50, VWP125, or VWP200)[1]

Waiting period	Interval length[2]	Range	SEM
VWP50	363^a	43–565	12.2
VWP125	445^b	203–586	12.8
VWP200	481^b	69–575	12.5

[a,b]Different superscript letters indicate a difference among LSM within the column (a-b: $P < 0.01$; b-b: $P = 0.14$).
[1]Values represent LSM, range, and SEM.
[2]Calving interval or interval from calving until the cow left the experiment.

3.1 Lactation yield per day of calving interval

The VWP did not affect the milk yield per day of CInt for primiparous or multiparous cows (Table 3). The VWP did not affect FPCM yield per day of CInt for primiparous cows, whereas FPCM yield per day of CInt was higher for multiparous cows in VWP50 compared with multiparous cows in VWP200. When the relative yield in the first 6 wk was included in the model as a covariate, FPCM yield per day of CInt tended to be higher for multiparous cows in VWP50 compared with multiparous cows in VWP200. When only cows that had a second calf were included, FPCM yield per day of CInt was higher for multiparous cows in VWP50 compared with multiparous cows in VWP200, both with and without correction for the relative yield.

The VWP did not affect the protein or lactose yield per day of CInt. The fat yield per day of CInt was greater for multiparous cows compared with primiparous cows (1.18 vs. 1.03 kg/d, $P < 0.01$). The protein yield per day of CInt was greater for multiparous cows compared with primiparous cows (1.00 vs. 0.86 kg/d, $P < 0.01$). The lactose yield per day of CInt was greater for multiparous cows compared with primiparous cows (1.24 vs. 1.07 kg/d, $P < 0.01$).

Table 3. Lactation yield per day of CInt[1], fat, protein, and lactose yield per day of CInt[2], and lactation persistency[3] of primiparous and multiparous cows with a voluntary waiting period after calving until first insemination of 50, 125, or 200 d (VWP50, VWP125, or VWP200), LSM ± SEM; all variables are expressed in kg/d of CInt unless stated otherwise

Item	Primiparous cows (n = 41)			SEM	Multiparous cows (n = 113)			SEM	*P*-value[4]		
	VWP50	VWP125	VWP200		VWP50	VWP125	VWP200		VWP	Par	VWP×Par
n	14	15	12		40	34	39				
Milk	23.3	22.7	23.5	1.45	29.5	28.0	25.6	0.86	0.26	<0.01	0.18
Milk, corrected[5]	23.1	22.7	23.8	1.09	28.8	27.9	26.4	0.65	0.58	<0.01	0.17
Milk, excluding culled and np[6] (n = 127)	23.1	22.8	23.9	1.50	28.8	27.3	24.8	0.85	0.37	<0.01	0.10
Milk, corrected, excluding culled and np (n = 127)	22.9	22.9	24.6	1.03	28.0^{a}	27.4^{ab}	25.4^{b}	0.58	0.83	<0.01	0.02
FPCM	24.5	24.5	25.7	1.31	30.4^{a}	29.5^{ab}	27.2^{b}	0.78	0.64	<0.01	0.09
FPCM, corrected[7]	24.2	24.5	25.9	1.03	30.0†	29.2	27.9†	0.61	0.96	<0.01	0.06
FPCM, excluding culled and np (n = 127)	24.4	24.6	25.6	1.39	29.7^{a}	28.8^{ab}	26.4^{b}	0.79	0.58	<0.01	0.09
FPCM, corrected, excluding culled and np (n = 127)	24.2	24.7	26.4	1.02	29.3^{a}	28.6^{ab}	26.8^{b}	0.58	0.99	<0.01	<0.01
Fat	0.99	1.02	1.08	0.05	1.22	1.21	1.12	0.03	0.93	<0.01	0.07
Protein	0.86	0.84	0.87	0.04	1.05	1.01	0.94	0.03	0.33	<0.01	0.15
Lactose	1.07	1.04	1.08	0.07	1.33	1.25	1.14	0.04	0.22	<0.01	0.17
Persistency, d 100–200 (kg/d)	−0.04	−0.04	−0.04	0.01	−0.10	−0.09	−0.07	0.01	0.52	<0.01	0.48
Persistency, d 100 to dry (kg/d)	−0.05	−0.04	−0.04	0.01	−0.10	−0.09	−0.07	0.01	0.03	<0.01	0.47

[a,b] Different superscript letters indicate a difference among LSM within the row and within the parity class ($P < 0.05$).

[1] CInt = calving interval. Milk yield in the first lactation within the experiment per day of calving interval or per day until culling (kg of milk or kg of fat- and protein-corrected milk (FPCM) per day).

[2] Effective yield of fat, protein, and lactose in the first lactation within the experiment per day of calving interval or per day until culling (kg/d).

[3] Reduction in milk yield in the first lactation within the experiment (kg of milk/d).

[4] VWP = voluntary waiting period. Par = parity class (primiparous or multiparous).

[5] Including relative milk yield in the first 6 wk of lactation in the model as covariate, $P < 0.01$.

[6] np = nonpregnant, cows that did not conceive during the first lactation of the experiment.

[7] Including relative FPCM yield in the first 6 wk of lactation in the model as covariate, $P < 0.01$.

† Trend in difference among LSM within the row and within the parity class ($P < 0.10$).

3.2 Lactation persistency

The VWP did not affect the persistency between d 100 and 200 of the lactation. Between d 100 and the start of dry-off, cows in VWP200 were more persistent compared with cows in VWP50 (−0.05 vs. −0.07 kg/d, $P = 0.02$). Primiparous cows were more persistent compared with multiparous cows between d 100 and 200 (−0.04 vs. −0.09 kg/d, $P < 0.01$) and between d 100 and the start of dry-off (−0.04 vs. −0.08 kg/d, $P < 0.01$).

For multiparous cows in VWP125 and VWP200, we detected a correlation between persistency between d 100 and 200 and persistency between d 100 and the start of dry-off (VWP125: 0.56, $P < 0.01$; VWP200: 0.74, $P < 0.01$). For primiparous cows in VWP50, we detected a trend for correlation between the 2 persistency measures (0.51, $P = 0.07$). For the other VWP × parity class combinations no correlation between the persistency measures was detectable. Overall, in VWP200 correlation between the 2 persistency measures was strongest (0.74, $P < 0.01$), followed by VWP125 (0.69, $P < 0.01$), and VWP50 (0.44, $P < 0.01$).

3.3 Milk yield before dry-off

During the last 6 wk before dry-off, after different VWP, VWP affected milk and FPCM yield, where cows in VWP50 had greater yield compared with VWP125 (milk: 18.9 ± 0.74 vs. 16.0 ± 0.75 kg/d, $P = 0.02$; FPCM: 22.1 ± 0.83 vs. 19.3 ± 0.84 kg/d, $P = 0.047$) and tended to have greater yield compared with VWP200 (milk: 18.9 ± 0.74 vs. 16.1 ± 0.90 kg/d, $P = 0.05$; FPCM: 22.1 ± 0.83 vs. 19.2 ± 1.01 kg/d, $P = 0.08$). At the moment of dry-off, milk yield was greater for cows in VWP50 compared with cows in VWP125 (18.2 ± 0.89 vs. 14.5 ± 0.91 kg/d, $P = 0.01$), and both VWP50 and VWP125 did not differ from VWP200 (15.4 ± 1.10 kg/d).

3.4 Weekly milk yield and fat, protein, and lactose content

During the first 44 wk of lactation, effect of VWP on milk yield and FPCM depended on week in lactation, and effect on milk yield tended to depend on parity (Table 4). Milk yield tended to be higher for multiparous cows in VWP125 compared with multiparous cows in VWP200. When the relative yield in the first 6 wk was included in the model, no differences in milk or FPCM yield were detectable among the VWP × parity classes, but FPCM yield tended to be higher for cows in VWP125 compared with cows in VWP50 (31.1 vs. 29.9 kg/d, $P = 0.08$). When only cows that had a second calf were included and relative yield was included as a covariate, primiparous cows in VWP200 had greater milk and FPCM yield compared with primiparous cows in VWP50. In this model, cows in VWP200 tended to have greater yield compared with cows in VWP50 (milk: 30.5 vs. 29.1 kg/d, $P = 0.08$; FPCM: 31.5 vs. 30.3 kg/d, $P = 0.08$).

The VWP did not affect fat, protein, or lactose content in the first 44 wk of lactation. Parity class did not affect fat or protein content. The lactose content was greater for primiparous cows compared with multiparous cows (4.60 vs. 4.47%, $P < 0.01$).

During the first 6 wk of the second lactation within the experiment, cows in VWP50 tended to have greater milk yield compared with cows in VWP200 (37.4 vs. 33.7 kg/d, $P = 0.05$). No differences were detectable among the VWP groups in FPCM in the first 6 wk of the second lactation.

3.5 Weekly body weight and monthly body condition score

Voluntary waiting period did not affect BW or BCS during the first 44 wk of the lactation. During the last 12 wk before dry-off, multiparous cows in VWP200 had higher BCS compared with multiparous cows in VWP50 and VWP125 (Table 5). The VWP did not affect BW or BCS in late lactation of primiparous cows or in their subsequent lactation. For multiparous cows, during the first 6 wk of the second lactation within the experiment, VWP200 resulted in a higher BCS compared with VWP50 and VWP125.

During the first 44 wk of lactation, primiparous cows had lower BW compared with multiparous cows (588 vs. 693, $P < 0.01$) and higher BCS compared with multiparous cows (2.5 vs. 2.3, $P < 0.01$). During the last 12 wk before dry-off, primiparous cows had lower BW compared with multiparous cows (663 vs. 756 kg, $P < 0.01$).

Table 4. Mean yield (kg of milk/d or kg of FPCM[1]/d) for the first 6 wk in lactation, the first 44 wk in lactation, 6 wk before dry-off, and 6 wk into the next lactation, of primiparous and multiparous cows with a voluntary waiting period after calving until first insemination of 50, 125, or 200 d (VWP50, VWP125, or VWP200); LSM ± SEM

Item	Primiparous cows (n = 41)				Multiparous cows (n = 113)				*P*-value[2]				
	VWP50	VWP125	VWP200	SEM	VWP50	VWP125	VWP200	SEM	VWP	Par	VWP× Par	Week× VWP	Week× Par
Yield in the first 6 wk in lactation													
Milk	26.1	25.5	25.5	1.67	39.0	37.9	36.1	1.00	0.41	<0.01	0.64	0.61	<0.01
FPCM	27.1	26.1	25.7	1.66	40.2	40.1	37.9	1.00	0.35	<0.01	0.80	0.99	<0.01
Yield in first 44 wk in lactation													
Milk	25.2	26.2	26.9	1.25	33.2	33.7†	31.0†	0.76	0.56	<0.01	0.09	<0.01	<0.01
Milk, corrected[3]	25.1	26.3	27.3	0.75	32.4	33.6	32.1	0.45	0.10	<0.01	0.05	<0.01	<0.01
Milk, excluding culled and np[4] (n = 127)	25.7	26.6	27.7	1.45	33.6	33.6	31.8	0.84	0.88	<0.01	0.19	<0.01	<0.01
Milk, corrected, excluding culled and np	25.5^{a}	26.7^{ab}	28.5^{b}	0.85	32.6	33.7	32.5	0.49	0.06	<0.01	0.03	<0.01	<0.01
FPCM	26.6	27.6	28.0	1.08	34.0	34.9	32.3	0.65	0.41	<0.01	0.12	<0.01	<0.01
FPCM, corrected[5]	26.5	27.6	28.5	0.64	33.4	34.5	33.4	0.39	0.05	<0.01	0.06	<0.01	<0.01
FPCM, excluding culled and np (n = 127)	27.0	27.9	28.4	1.26	34.3	34.7	32.9	0.73	0.70	<0.01	0.31	<0.01	<0.01
FPCM, corrected, excluding culled and np	26.8^{a}	28.0^{ab}	29.5^{b}	0.75	33.7	34.5	33.5	0.43	0.05	<0.01	0.02	<0.01	<0.01
Content in the first 44 wk in lactation													
Fat (%)	4.37	4.40	4.32	0.08	4.22	4.29	4.32	0.05	0.70	0.09	0.43	0.56	0.37
Protein (%)	3.73	3.64	3.60	0.06	3.66	3.61	3.66	0.04	0.28	0.74	0.34	0.98	0.52
Lactose (%)	4.59	4.59	4.61	0.02	4.50	4.46	4.45	0.01	0.58	<0.01	0.12	0.95	0.98
Yield in the last 6 wk before dry-off													
Milk	19.8	17.2	19.3	1.59	18.0	14.9	12.8	0.89	0.01	<0.01	0.10	<0.01	<0.01
FPCM	23.5	21.0	23.2	1.78	20.8	17.5	15.2	0.99	0.02	<0.01	0.10	<0.01	<0.01
Yield in the first 6 wk next lactation													
Milk	36.1	34.4	32.1	2.03	38.8	34.7	35.3	1.21	0.03	0.09	0.55	0.08	0.01
FPCM	41.8	39.2	36.9	2.15	42.5	40.2	40.7	1.30	0.11	0.16	0.59	<0.01	0.15

[a,b] Different superscript letters indicate a difference among LSM within the row and within the parity class ($P < 0.05$). Week had *P*-value < 0.01 in all analyses.

[1] FPCM = fat- and protein-corrected milk (CVB, 2012).

[2] VWP = voluntary waiting period. Par = parity class (primiparous or multiparous).

[3] Including relative milk yield in the first 6 wk of lactation in the model as covariate, $P < 0.01$.

[4] np = nonpregnant, cows that did not conceive during the first lactation of the experiment.

[5] Including relative FPCM yield in the first 6 wk of lactation in the model as covariate, $P < 0.01$.

[†] Trend in difference among LSM within the row and within the parity class ($P < 0.10$).

Table 5. Mean BW (kg) and mean BCS for the first 6 wk in lactation, the first 44 wk in lactation, 6 wk before dry-off, and 6 wk into the next lactation, of primiparous and multiparous cows with a voluntary waiting period after calving until first insemination of 50, 125, or 200 d (VWP50, VWP125, or VWP200); LSM ± SEM

Item	Primiparous cows (n = 41)				Multiparous cows (n = 113)				*P*-value[1]				
	VWP50	VWP125	VWP200	SEM	VWP50	VWP125	VWP200	SEM	VWP	Par	VWP× Par	Time× VWP	Time× Par
First 6 wk in lactation													
BW	549	543	558	9.4	692	693	680	5.7	0.98	<0.01	0.57	0.65	0.72
BCS (first 1.5 mo)[2]	2.7	2.6	2.4	0.09	2.3	2.4	2.3	0.06	0.35	0.06	0.66	0.58	0.91
First 44 wk in lactation													
BW	592	578	592	8.4	700	692	687	5.1	0.63	<0.01	0.71	0.78	0.49
BCS (first 11 mo)	2.6	2.6	2.4	0.05	2.3	2.3	2.4	0.03	0.39	<0.01	0.19	0.40	0.14
Last 12 wk before dry-off													
BW	655	661	672	9.5	737	752	779	5.8	0.10	<0.01	0.67	0.39	0.93
BCS (last 3 mo)	2.5	2.9	2.7	0.09	$2.6^{a\dagger}$	$3.0^{a\dagger}$	3.5^{b}	0.06	<0.01	<0.01	0.01	0.61	0.20
First 6 wk in next lactation													
BW	637	627	647	11.1	701	694	721	7.0	0.36	<0.01	0.95	0.65	0.99
BCS (first 1.5 mo)[2]	2.5	2.9	2.8	0.10	2.7^{a}	3.0^{a}	3.5^{b}	0.06	<0.01	0.01	0.08	0.58	0.96

[a,b] Different superscript indicates a difference among LSM within the row and within the parity class ($P < 0.05$). Week had *P*-value < 0.01 in all analyses. Month had *P*-values as follows: $P < 0.01$ for the first 1.5 mo in lactation, $P = 0.09$ for the first 11 mo in lactation, $P < 0.01$ for the last 3 mo before dry-off, and $P = 0.03$ for the first 1.5 mo of the next lactation within the experiment.

[1] VWP = voluntary waiting period. Par = parity class (primiparous or multiparous). Time represents week (BW) or month (BCS).

[2] For some cows in this time period, 1 evaluation of BCS occurred; for others in this time period, 2 evaluations of BCS occurred.

[†] Trend in difference among LSM within the row and within the parity class ($P < 0.10$).

3.6 Cow characteristics as predictors for lactation performance

After backward selection, the final multivariate model for FPCM yield per day of CInt included VWP class and parity class as class variables, and FPCM, milk yield, BW, interaction of BW × parity class, maximum yield, interaction maximum yield × VWP, slope to maximum yield, interaction of slope to maximum yield × VWP, expected (primiparous cows) or previous (multiparous cows) 305-d milk yield, breeding value for persistency, and interaction of breeding value for persistency × VWP as continuous variables (Table 6). In this model, maximum yield in the first 6 wk of lactation was positively associated with FPCM per day of CInt in all VWP groups, most in VWP50. Slope to maximum yield was positively associated with FPCM per day of CInt in VWP125 and negatively associated with FPCM per day of CInt in VWP50 and VWP200. The breeding value for persistency was positively associated with FPCM per day of CInt in all VWP groups, most in VWP200. Mean BW in the first 6 wk of lactation was positively associated with FPCM per day of CInt for primiparous cows and negatively associated with FPCM per day of CInt for multiparous cows. Mean FPCM in the first 6 wk of lactation was positively associated with FPCM per day of CInt, whereas mean milk yield in the first 6 wk of lactation was negatively associated with FPCM per day of CInt.

After backward selection, the final multivariate model for lactation persistency from d 100 until the start of dry-off included VWP class and parity class as class variables, and milk yield, interaction of milk yield × VWP, maximum yield, interaction of maximum yield × VWP, and expected (primiparous cows) or previous (multiparous cows) 305-d yield as continuous variables (Table 7). In this model, mean milk yield in the first 6 wk of lactation was negatively associated with lactation persistency in VWP125 and VWP200, and positively associated with lactation persistency in VWP50. Maximum milk yield in the first 6 wk of lactation was negatively associated with lactation persistency in VWP50 and positively associated with lactation persistency in VWP125 and VWP200. Expected or previous 305-d yield was negatively associated with lactation persistency.

Table 6. Final multivariable model for prediction of fat- and protein-corrected milk yield (FPCM) per day of calving interval (in kg/d) for cows with a voluntary waiting period (VWP) after calving until first insemination of 50, 125, or 200 d (VWP50, VWP125, or VWP200); LSM ± SEM or regression coefficient (β) with SE and range[1]

Variable	Category	LSM (SEM) or *β (SE)*	Range[2]	*P*-value
VWP	50	30.1^a (0.79)		0.02
	125	30.1^a (0.82)		
	200	28.8^b (0.77)		
Parity	1	31.8 (0.59)		0.06
	2+	27.6 (0.34)		
FPCM[3]		*0.573 (0.113)*	18.8–55.5	<0.01
Milk yield[3]		*−0.844 (0.220)*	17.2–51.9	<0.01
BW[3]		*0.019 (0.014)*	493–870	0.57
BW × Parity	1[4]	*0 (−)*	493–646	0.04
	2+	*−0.030 (0.014)*	526–870	
Maximum yield[3]		*0.835 (0.210)*	21.2–59.9	<0.01
Maximum yield × VWP	50[4]	*0 (−)*	22.0–59.9	<0.01
	125	*−0.283 (0.068)*	24.0–58.8	
	200	*−0.249 (0.066)*	21.2–57.8	
Slope to maximum yield[3]		*−1.802 (1.114)*	0.14–1.92	0.93
Slope × VWP	50[4]	*0 (−)*	0.14–1.04	<0.01
	125	*4.686 (1.481)*	0.15–0.88	
	200	*0.909 (1.331)*	0.18–1.92	
305-d milk yield[5]		*0.568 (0.243) × 10−3*	5,862–14,343	0.02
Breeding value persistency		*0.232 (0.127)*	92–113	<0.01
Breeding value persistency × VWP	50[4]	*0 (−)*	95–108	0.03
	125	*−0.040 (0.152)*	92–113	
	200	*0.322 (0.169)*	92–108	

[a,b] Different superscript letters indicate a difference among LSM within the column within 1 variable ($P < 0.05$).

[1] The final multivariate model was based on 14 univariate models, with individual early-lactation variables as independent variable, to identify potential predictors for milk yield after different VWP.

[2] Ranges for FPCM, milk yield, maximum yield, and slope to maximum yield in kg/d; ranges for BW and 305-d milk yield in kg.

[3] Measured in the first 6 wk after the first calving within the experiment.

[4] Reference category.

[5] Expected (primiparous cows) or previous (multiparous cows) 305-d milk yield.

Table 7. Final multivariable model for prediction of lactation persistency (in kg/d) between d 100 and start of dry-off for cows with a voluntary waiting period (VWP) after calving until first insemination of 50, 125, or 200 d (VWP50, VWP125, or VWP200); LSM ± SEM or regression coefficient (β) with SE and range[1]

Variable	Category	LSM (SEM) or *β (SE)*	Range[2]	*P*-value
VWP	50	−0.077 (0.003)		0.37
	125	−0.071 (0.004)		
	200	−0.059 (0.004)		
Parity	1	−0.059 (0.005)		<0.01
	2+	−0.078 (0.003)		
Milk yield[3]		*0.003 (0.002)*	17.2–51.9	0.06
Milk yield × VWP	50[4]	*0 (−)*	17.7–51.9	<0.01
	125	*−0.008 (0.004)*	19.8–49.1	
	200	*−0.011 (0.003)*	17.2–48.7	
Maximum yield[3]		*−0.004 (0.002)*	21.2–59.9	0.47
Maximum yield × VWP	50[4]	*0 (−)*	22.0–59.9	<0.01
	125	*0.006 (0.003)*	24.0–58.8	
	200	*0.010 (0.003)*	21.2–57.8	
305-d milk yield		*−0.455 (0.207) × 10−5*	5,862–14,343	0.03

[a,b] Different superscript letters indicate a difference among LSM within the column within 1 variable ($P < 0.05$).

[1] The final multivariate model was based on 14 univariate models, with individual early-lactation variables as independent variable, to identify potential predictors for lactation persistency after different VWP.

[2] Ranges for milk yield and maximum yield in kg/d; range for 305-d milk yield in kg.

[3] Measured in the first 6 wk after the first calving within the experiment.

[4] Reference category.

4 Discussion

Increasing VWP until 200 d did not affect milk or FPCM yield per day of CInt for primiparous cows. Primiparous cows seem to be appropriate to select for an extended VWP, without losing milk. This is comparable to other studies, where primiparous cows kept for extended lactations achieved similar or even greater lactation yields compared with primiparous cows in shorter lactations (Arbel et al., 2001; Lehmann et al., 2016). The main reason that primiparous cows achieve the same yield per day of CInt after an extended VWP as after a short VWP of 50 d is their high lactation persistency (Arbel et al., 2001; Lehmann et al., 2016; Niozas et al., 2019a). Also in the current study, primiparous cows had greater lactation persistency compared with multiparous cows. This higher persistency of primiparous cows compared with multiparous cows possibly also resulted in their higher yield in the last 6 wk before and at dry-off in the current study. When primiparous cows are kept for an extended CInt, however, they take more time to become a second-parity cow. Second-parity cows are, in general, more productive than primiparous cows (Friggens et al., 1999; Lee and Kim, 2006). Therefore, when primiparous cows were kept for an extended CInt, this still resulted in a loss in milk yield at herd level (Kok et al., 2019).

Increasing VWP until 125 d did not affect milk or FPCM yield per day of CInt for multiparous cows. When extending VWP further, until 200 d, the yield per day of CInt was lower compared with VWP50. This difference was around 2 to 4 kg/d, depending on whether culled and nonpregnant cows were included or whether a correction for the relative yield was used. The reason for the lower production of multiparous cows in VWP200 could be that they have more days at the end of this long lactation, where milk yield is usually lower. Also, in our earlier study at commercial farms, cows with the greatest production potential that had calving intervals >531 d could not sustain production in these long CInt (Burgers et al., 2021a). In that study, however, the long CInt consisted of both cows with a voluntarily extended waiting period for insemination and cows that failed to conceive at earlier insemination(s). In another study, milk yield per day of CInt did not decrease when calving interval was extended up to 18 mo (Österman and Bertilsson, 2003). In that study, some of the cows were milked 3 times per day, increasing their peak milk yield and their persistency, resulting in similar productions compared with a calving interval of 12 mo. In line with other studies on deliberately extended lactations (Arbel et al., 2001; Lehmann et al., 2016), we used yield per day of CInt to compare milk yield after different VWP. Moreover, yield per day of CInt is economically of interest: extended lactations can be profitable when yield per day of CInt is maintained at similar levels as in shorter CInt, or when milk yield losses can be compensated for by lower costs, as for insemination, feed, or disease.

At the moment of dry-off, milk yield was lower for cows in VWP125 compared with cows in VWP50. Yield for cows in VWP200 was not significantly lower at dry-off compared with cows in VWP125 or VWP50. This could be explained by the greater lactation persistency between d 100 and the start of dry-off of cows with a VWP of 200 d compared with cows after a VWP of 50 d, possibly related to a delayed effect of pregnancy on the lactation curve due to later gestation (Strandberg and Lundberg, 1991; Kok et al., 2019). Although milk yield decreased in late lactation, fat and protein content in milk increased toward late lactation, as it did in an earlier study (Silvestre et al., 2009). The total solid yield, however, did not increase in our study, as the total production of milk was lower in late lactation, especially after extended VWP. In the current study, an extended VWP relative to a VWP of 50 d resulted in a lower milk yield before dry-off, which could benefit udder health in the dry period and at calving (Rajala-Schultz et al., 2005; Odensten et al., 2007). The lower milk yield before dry-off could be related to an increased risk for fattening of cows in late lactation. The BCS of cows in VWP200 and VWP125 were 3.1 and 2.9 during the last 3 mo before dry-off, compared with a BCS of 2.6 for cows in VWP50. This greater BCS could increase the risk for diseases after next calving (Roche and Berry, 2006).

During the first 44 wk in lactation, we detected no differences for FPCM yield among VWP groups. During the first 6 wk of the lactation where VWP treatment was applied, both FPCM and milk yield were numerically lower for multiparous cows with VWP200 compared with multiparous cows with VWP50, and VWP125 had a yield in between. During the first 6 wk, no effect of VWP is possible, and therefore we corrected the 305-d milk yield for the yield in these first 6 wk. After this correction, and only including cows that had a second calf within the experiment, primiparous cows in VWP200 had greater 305-d yield compared with primiparous cows in VWP50. During the last 50 d of the first 305 d, pregnancy could already affect the lactation curves of some cows in VWP50 (Strandberg and Lundberg, 1991; Kok et al., 2019). This pregnancy effect may explain the somewhat lower 305-d yield in VWP50.

During the first 6 wk of the second lactation within the experiment, cows in VWP50 tended to have greater milk yield compared with cows in longer VWP, but FPCM yield was the same in the 3 VWP groups. Although primiparous cows from VWP200 did have more time to grow before their second calving compared with primiparous cows from VWP50, they did not achieve greater milk yield or FPCM during the first 6 wk after the second calving. In an earlier study, the increase in milk production in the subsequent lactation compared with the previous lactation was greater for primiparous and multiparous cows that had 2 subsequent lactations of 18 mo compared with 2 subsequent lactations of 12 mo (Österman and Bertilsson, 2003). In observational data from commercial farms, second-parity cows achieved greater ECM per day of CInt when their previous CInt was extended compared with when their previous CInt was shorter (Lehmann et al., 2016). In the current experiment, we monitored only the first 42 d of the next lactation. This may be too short a time period for cows to show their possible higher production potential. Another reason for the tendency for lower milk yield in VWP200 could be that only 9 out of 12 primiparous cows in VWP200 had a second calf, which is a relatively low number, also compared with the primiparous cows in VWP125 (14 out of 15) and in VWP50 (13 out of 14).

Fat- and protein-corrected milk yield per day of CInt of cows with different VWP could be predicted by the maximum yield in the first 6 wk of lactation, the slope to this maximum yield, and the breeding value for persistency. In the model, the breeding value for persistency had a more positive relation with FPCM per day of CInt in VWP200, with VWP50 as a reference value. This could indicate that cows with a greater breeding value for persistency perform better in VWP200 compared with VWP50 in terms of FPCM yield per day of CInt, possibly related to the higher importance of lactation persistency for total milk yield after longer VWP for insemination compared with a shorter VWP (Lehmann et al., 2016; Kok et al., 2019). Moreover, in the model, maximum yield had a less positive effect on FPCM per day of CInt in the longer VWP groups compared with VWP50, possibly related to the lower lactation persistency that is related to a greater peak yield (Dekkers et al., 1998).

Lactation persistency between d 100 and moment of dry-off of cows with different VWP could be predicted by mean milk yield and maximum yield in the first 6 wk. In VWP50, mean milk yield had a positive relation to lactation persistency, and maximum yield had a negative relation to lactation persistency. In VWP125 and VWP200, these effects were reversed compared with VWP50. A higher peak yield is often associated with lower persistency (Dekkers et al., 1998). This can be related to the more negative energy balance when peak yield is higher, which could be related to reduced persistency in later lactation (Chen et al., 2016). Next to these early-lactation production characteristics, the expected (primiparous cows) or previous (multiparous cows) 305-d yield was also related to lactation persistency between d 100 and the start of dry-off. A greater 305-d yield was related to lower persistency, possibly because a greater 305-d yield is often related to a greater peak yield, which is related to decreased persistency (Dekkers et al., 1998; Chen et al., 2016).

In the current study, we investigated the effects of 3 VWP on milk yield and lactation persistency in a controlled experiment, in contrast with the work on extended lactations that is performed in observational studies on farms (e.g., Lehmann et al., 2016; Mellado et al., 2016; Burgers et al., 2021a). All cows were blocked and randomly assigned to one of the 3 VWP groups, which had a fixed VWP in days, making it possible to find cow characteristics that contribute to milk yield of individual cows in different lactation lengths. Some of the cow characteristics found in this study that affected total lactation yield or persistency after different VWP might be additionally used by farmers to optimize VWP for individual cows. Selecting specific cows for extended VWP could imply that herd-level benefits of longer CInt, such as reduced frequency of transitions such as dry-off and calving, reduced labor related to these transitions and reduced the number of surplus calves, as well as minimizing loss of milk yield on a herd level. Some farmers already used different cow characteristics in early lactation to determine for which cows they extended the VWP (Lehmann et al., 2016; Burgers et al., 2021a). Early-lactation characteristics that were used in our earlier study included maximum yield, BCS, and BW (Burgers et al., 2021a). In practice, farmers often extended VWP until cows reached a certain milk level (Burgers et al., 2021a). Waiting until milk drops below a certain level helps in selecting more persistent cows for longer CInt, as the longer a cow takes to reach this milk level, the longer the VWP will be, and often the more persistent this cow is. Together with the early-lactation cow characteristics, a waiting period based on milk level might contribute to an individual approach for extended VWP management.

5 Conclusions

For both primiparous and multiparous cows, VWP was extended until 125 d with no effect on milk or FPCM yield per day of CInt. For primiparous cows, extending the VWP further until 200 d still did not affect yield per day of CInt, although, for multiparous cows, extending the VWP to 200 d resulted in a lower yield per day of CInt. Moreover, cows in longer VWP had lower yield at dry-off, which may benefit their udder health during the dry period and possibly also the subsequent lactation. On the contrary, multiparous cows in longer VWP had higher BCS at dry-off and in the first weeks of the subsequent lactation, which may hamper metabolic health and adaptation to a new lactation. Milk characteristics in the first 6 wk of lactation and the breeding value for persistency determined cow performance after different VWP.

6 Acknowledgments

The authors thank the staff of the Dairy Campus (Leeuwarden, the Netherlands). This study was financed by DairyNL (ZuivelNL; organization of the Dutch dairy supply chain) and the Dutch Ministry of Agriculture, Nature and Food Quality (LNV) as part of the research program One Health for Food (1H4F). The authors have not stated any conflicts of interest.

Appendix

Table A1. Mean range within blocks of the variables used to select cows for blocks: calving date (d), expected (primiparous cows) or previous (multiparous cows) 305-d milk yield (kg), parity, and breeding value for persistency

Variable	Mean range
Calving date	26
305-d milk yield	975
Parity	0.6
Breeding value persistency	4.9

Table A2. Milk and fat- and protein-corrected milk yield (FPCM) per day of calving interval (CInt)[1], and lactation persistency[2] of primiparous and multiparous cows with CInt of <415 d (CInt-1), 415 to 485 d (CInt-2), or >485 d (CInt-3)

Item	Primiparous cows (n = 36)			SEM	Multiparous cows (n = 91)			SEM	*P*-value[3]		
	CInt-1	CInt-2	CInt-3		CInt-1	CInt-2	CInt-3		CInt	Par	CInt×Par
n	12	13	11		31	31	29				
Milk	23.9	21.9	23.9	1.37	28.7	27.1	25.3	0.84	0.18	<0.01	0.17
FPCM	25.1	23.9	25.5	1.26	29.8	28.4	26.8	0.78	0.33	<0.01	0.19
Persistency d 100 – dry[4]	−0.04	−0.04	−0.04	0.01	−0.10	−0.09	−0.07	0.01	0.11	<0.01	0.40

[1] Milk yield in the first lactation within the experiment per day of calving interval (kg of milk or kg of FPCM per day).
[2] Reduction in milk yield in the first lactation within the experiment (kg of milk/d).
[3] Par = parity class (primiparous or multiparous).
[4] Persistency between d 100 of lactation and dry-off.

Chapter 4

Effect of voluntary waiting period on metabolism of dairy cows during different phases of the lactation

E.E.A. Burgers[1,2], R.M.A. Goselink[2], R.M. Bruckmaier[3], J.J. Gross[3], R. Jorritsma[4], B. Kemp[1], A. Kok[1], A.T.M. van Knegsel[1]

[1]Adaptation Physiology Group, Wageningen University & Research, Wageningen, the Netherlands
[2]Wageningen Livestock Research, Wageningen University & Research, Wageningen, the Netherlands
[3]Veterinary Physiology, Vetsuisse Faculty, University of Bern, Bern, Switzerland
[4]Department of Farm Animal Health, Ruminant Health Unit, Utrecht University, Utrecht, the Netherlands

Submitted

Abstract

An extended calving interval (CInt) by extending the voluntary waiting period (VWP) could be associated with altered metabolic status and energy partitioning in dairy cows, due to delayed gestation and a longer period in late lactation with relatively lower milk production. The aim of this study was first to evaluate the effects of the VWP on metabolic status, body condition, and energy partitioning during different phases of the lactation: during the first 305 days after the first calving in the experiment (calving 1), around the end of the VWP, around successful insemination, and during the pregnancy period (i.e., period of 280 d before calving 2). Second, the effects of the VWP and CInt on metabolic status during the next transition period were determined from 2 wk before until 6 wk after calving 2. Third, individual cow characteristics as monitored before successful insemination were used to predict lactation performance of cows after different VWP. Holstein-Friesian cows (N=153) were blocked and randomly assigned to a VWP of 50, 125, or 200 days (VWP50, VWP125, or VWP200), and followed from 2 wk before calving 1 until 6 wk after calving 2. Weekly, from 2 wk before until 6 wk after calving, plasma samples were analyzed for non-esterified fatty acids (NEFA), β-hydroxybutyrate, glucose, insulin and insulin-like growth factor 1 (IGF-1). During lactation, plasma samples were analyzed for insulin and IGF-1 every 2 wk, and FPCM and BW gain were calculated weekly. Cows were divided in two parity classes based on calving 1 (primiparous and multiparous) and remained in these classes after calving 2. Around the end of the VWP, multiparous cows in VWP200 had greater plasma insulin (17.6 μU/mL) and IGF-1 concentration and lower fat- and protein-corrected milk (FPCM) production (30.1 kg/d) compared with cows in VWP125 (insulin: 13.7 μU/mL; FPCM: 36.9 kg/d) or VWP50 (insulin: 13.4 μU/mL; FPCM: 42.4 kg/d). During the pregnancy period, multiparous cows in VWP200 had greater plasma insulin and IGF-1 concentration and lower FPCM production compared with cows in VWP50 or VWP125 and had greater daily BW gain compared with cows in VWP50 (3.6 vs. 2.5 kg/d). For primiparous cows, the VWP did not affect plasma hormones, FPCM production, or body condition around the end of the VWP. During the pregnancy period, primiparous cows in VWP125 had greater plasma insulin concentration compared with primiparous cows in VWP50, but the VWP never affected body condition or FPCM production for primiparous cows. In the first 6 wk after calving 2, multiparous cows in VWP200 had greater plasma NEFA concentration compared with multiparous cows in VWP125 or VWP50 (0.41 vs. 0.30 or 0.26 mmol/L). For primiparous cows, the VWP did not affect the metabolism, BW, or FPCM production during the 6 wk after calving 2. Independent of the VWP, higher milk production and lower body condition before insemination were associated with higher milk production and lower body condition score end lactation. Variation in these characteristics among cows could call for an individual approach for an extended VWP.

Key words: extended calving interval, extended lactation, energy partitioning, individual cow variation

1 Introduction

Extending the calving interval (CInt) by extending the voluntary waiting period (VWP) is a strategy to reduce the frequency of calving-related transitions. This strategy could thereby reduce the risk for diseases per unit of time, as most diseases are associated with the period around calving (Friggens et al., 2004). When the VWP is extended, insemination takes place at a later moment in lactation, when milk production is decreased (Gaillard et al., 2016). At that time, there will be less energy partitioned toward milk and more toward other body functions, which could improve fertility after an extended VWP (Wathes et al., 2007; Niozas et al., 2019b).

Moreover, when the VWP and thus the CInt is extended, the shape of the lactation curve can be expected to be different than with a traditional 365 d CInt. For example, cows with longer VWP had more persistent lactation curves (Niozas et al., 2019a; Burgers et al., 2021b). Additionally, when the VWP was extended, cows had a lower milk production and increased BCS at the end of lactation (Niozas et al., 2019a; Burgers et al., 2021b). Differences in energy partitioning between milk and body reserves can be hypothesized to be related with changes in hormone concentrations during lactation. A lower plasma insulin concentration was for example related to less energy partitioning toward body reserves and more toward milk (Hart, 1983). Moreover, lower insulin was related to lower inulin-like growth factor 1 (IGF-1) (Butler et al., 2003), which was associated with a higher GH concentration in rats and mice (Tannenbaum et al., 1983; Romero et al., 2012). For dairy cows, a higher GH concentration has been related to a higher milk production (Peel et al., 1983).

It can be hypothesized that after the extended lactation, cows with an extended VWP and related increased BCS at the end of the lactation could have a reduced feed intake and a more negative energy balance (EB) after the next calving. In that case, cows with an extended VWP in the previous lactation might have an increased risk for metabolic disorders after calving (Gillund et al., 2001; Roche and Berry, 2006), possibly reflected by an increased plasma concentration of non-esterified fatty acids (NEFA) and β-hydroxybutyrate (BHB) during the early phase of the lactation.

Not all cows with an extended VWP, however, have a reduced milk yield or an increased BCS at the end of the lactation, which could indicate that some cows might be more prone to partition energy toward milk rather than toward body reserves (Lehmann et al., 2016; Niozas et al., 2019a). For example, cows with a high milk production at 450 DIM (18.9 kg/d) had a greater tendency for lipid mobilization around 460 DIM and around 580 DIM compared with cows with a lower milk production at 450 DIM (12.3 kg/d) (Marett et al., 2019). Moreover, when cows had a lower plasma insulin and IGF-1 concentration between 332 and 612 DIM as a result of differences in diet composition, more cows were able to sustain milk production in a lactation of 612 days compared with cows with higher plasma concentration of insulin and IGF-1 between 332 and 612 DIM (Delany et al., 2010). In addition, individual cow characteristics, such as milk production and body weight in early lactation, were associated with milk production per day of CInt and lactation persistency in extended lactations (Burgers et al., 2021b). It can be hypothesized that milk production and fattening at the end of the lactation of cows with different VWP are related with milk production and BCS in early lactation, but also with insulin and IGF-1 concentration either in late lactation or already earlier in lactation. Early identification of cows that are able to maintain milk production with an extended VWP is beneficial for selection of cows for an extended VWP.

The aim of this study was first to evaluate the effects of 3 VWP (50, 125, and 200 d) on insulin, IGF-1, body condition, and energy partitioning between fat- and protein-corrected milk (FPCM) and BW during different phases of the lactation: during the first 305 days after the first calving in the experiment (calving 1), around the end of the VWP, around successful insemination, and during the pregnancy period (i.e., period of 280 d before calving 2). Second, the effects of the VWP and CInt on metabolic status during the next transition period were determined by analyzing body condition, dry matter intake (DMI), EB, plasma NEFA, BHB, and glucose concentration, and plasma insulin and IGF-1 concentration, from 2 wk before until 6 wk after calving 2. Third, individual cow characteristics as monitored before successful insemination were used to predict lactation performance of cows after different VWP. The 3 variables used as indicator for lactation performance were: 1. FPCM production during the final 6 wk before dry-off, 2. BCS during the final 12 wk before dry-off, and 3. FPCM production per day of CInt.

2 Materials and methods

2.1 Animals and housing

The experimental protocol was approved by the Institutional Animal Care and Use Committee of Wageningen University & Research (the Netherlands) and complied with the Dutch law on Animal Experimentation (protocol number 2016.D-0038.005). The experiment was conducted at Dairy Campus research farm (Leeuwarden, the Netherlands) between December 2017 and January 2020.

The animals, experimental design, and treatments have been described earlier (Burgers et al., 2021b). In short, 154 cows were selected from a research herd of 500 lactating Holstein Friesian cows based on the following criteria: no twin pregnancy, no clinical mastitis or SCC > 250,000 at the final 2 milk test days before dry-off and expected to finish a complete lactation based on being in good general health. For the current study, cows were followed from 2 wk before expected calving (calving 1) until 6 wk after calving 2 or until culling. Cows were milked twice daily around 6am and 6pm in a 40-cow rotary milking parlor (GEA, Dusseldorf, Germany). During lactation, cows were fed partially mixed ration (PMR): grass silage, corn silage, soybean meal, and wheat meal, supporting 22 kg of milk. Moreover, concentrate was supplied from the day of calving and increased until 21 DIM to 9 kg/d (primiparous cows) or 10 kg/d (multiparous cows). After 100 DIM, concentrate supply was decreased based on reductions in milk production. In the milking parlor cows additionally received 1 kg of concentrate per day. Ration during the dry period consisted of grass silage and corn silage, supplemented with wheat straw and concentrate. In the last 10 d before the expected calving date, cows received 1 kg concentrate daily. Once per week, cows between 42 and 49 days before the expected calving date were dried-off. In the 7 days prior to dry-off, cows were given the dry-cow ration. During the last 3 days before dry-off, cows were milked once daily. When cows had an SCC > 150,000 cells/mL at the final milk test day, cows were treated with antibiotics at dry-off (Orbenin Dry Cow Extra, Zoetis, the Netherlands). All cows were treated with teat sealant at dry-off (Orbeseal, Zoetis, the Netherlands).

2.2 Experimental design

The selected 154 animals were blocked for parity, calving date, milk production in the previous lactation (multiparous cows) or expected milk production (primiparous cows), and the breeding value for persistency (CRV, Arnhem, the Netherlands) in wk 6 after calving. Each block consisted of 3 cows. Per block, the cows were randomly divided over 3 treatment groups: a VWP of 50 days (VWP50), 125 days (VWP125), or 200 days (VWP200). Cows in the 3 treatment groups were inseminated at the first estrus after the end of their VWP. Estrus detection was carried out by using neck mounted 3D accelerometers (Nedap Smarttag Neck, Groenlo, the Netherlands) in combination with visual observations by the animal caretaker. Cows were inseminated until 300 DIM. Cows that did not conceive within 300 DIM stayed in the experiment until 530 DIM as long as they produced at least 10 L of milk/d.

2.3 Measurements and calculations

Milk production and body condition

Milk production was recorded at every milking. Milk samples were collected for each individual cow 4 times per week (Tuesday afternoon, Wednesday morning, Wednesday afternoon, Thursday morning) in 10 mL tubes containing Bronopol as a preservative and analyzed for the percentage of fat, protein, and lactose as a pooled sample [(ISO 9622, 2013), Qlip, Zutphen, the Netherlands)]. The FPCM was calculated as follows (CVB, 2016):

$$\mathrm{FPCM(kg)} = \mathrm{milk\ (kg)} \times (0.337 + 0.116 \times \mathrm{fat\ (\%)} + 0.06 \times \mathrm{protein\ (\%)}),$$

by using the weekly contents of fat and protein, and the mean daily milk production of each week. Body condition score was visually evaluated every 4 wk by the same person using a 1 to 5 scale (Ferguson et al., 1994). Body weight was measured twice daily after milking, by a scale that the cows walked over when returning from the milking rotary to the freestall (GEA, Dusseldorf, Germany). Dry cows were weighed once a week on the same scale. The difference in BW between 2 subsequent wk (ΔBW) was calculated by subtracting average BW in one wk from average BW in the next wk. For the calculation of ΔBW, week in which BW was averaged was shifted with 3 d, for ΔBW to follow the lactation weeks. The ΔBW was only calculated if BW was recorded at least 5 times in both weeks.

Energy balance

Weekly energy balance was calculated in the first 6 wk after calving 1, and from 2 wk before until 6 wk after calving 2, as the difference between intake of net energy (NE) and requirements of NE for maintenance, milk production, and pregnancy (CVB, 2016). To calculate intake of NE, daily intake of PMR was measured individually by using roughage intake control (RIC) troughs (Insentec, Marknesse, the Netherlands) with at least 1 trough available per 2 cows. Concentrate intake was recorded by concentrate feeders for all individual cows. The NE requirement for maintenance was assumed to be 291.18 kJ/BW0.75, NE requirement for milk production was assumed to be 3,049.8 kJ/kg FPCM, and NE requirement for pregnancy was assumed to be 18,630 kJ/d in the final 2 wk before calving. Energy intake, requirements, and EB are expressed in kJ/BW0.75 per day.

Energy partitioning

As DMI was not available for the complete lactation, the ratio of energy for FPCM to energy for maintenance was calculated to evaluate energy partitioning of cows during the lactation. Net energy for FPCM (NE-FPCM; in kJ, according to the Dutch net energy system for lactation) was calculated as 442 × FPCM (kg) (CVB, 2016). Net energy for maintenance (NE-BW) was calculated as 42.4 × BW (kg)0.75 (CVB, 2016). The ratio of NE-FPCM to NE-BW was calculated as NE-FPCM / NE-BW.

Blood collection and analysis

Blood was collected weekly in the first 6 wk after calving 1 in the experiment, and from 2 wk before until 6 wk after calving 2, for the analysis of plasma NEFA, BHB, glucose, insulin and IGF-1 concentration. From 7 wk after calving 1 until 2 wk before calving 2, blood was collected every 2 wk for the analysis of plasma insulin and IGF-1 concentration. After the morning milking, blood (10 mL) was collected from the coccygeal vessels into evacuated EDTA tubes (Vacuette, Greiner BioOne, Kremsmunster, Austria). Blood samples were kept on ice before centrifugation for plasma isolation (3,000 × *g* for 15 min, 4°C). Samples were stored at −20°C. Plasma insulin concentration was measured by using kit no. PI-12K from EMD Millipore Corporation (Billerica, MA, USA). Plasma IGF-1 concentration was measured by using kit no. A15729 from Beckman Coulter (Fullerton, CA, USA). Plasma metabolite concentration was measured by using an autoanalyzer (Cobas Mira, Roche), with the following enzymatic kits: NEFA: NEFA FA115; BHB: RANBUT RB1007; Glucose: GLUC-PAP GL364 (Randox Laboratories Ltd., Schwyz, Switzerland).

Cow characteristics as predictor for lactation performance

Lactation performance was defined with 3 variables: mean FPCM production in the final 6 wk before dry-off, mean BCS in the final 12 wk before dry-off, and FPCM production per day of CInt. Available individual cow characteristics used to predict lactation performance consisted of estimated 305-d milk production, the breeding value for persistency (CRV, Arnhem, the Netherlands), and cow characteristics in three periods: 1. The first 6 weeks of lactation, 2. Between calving and successful insemination, and 3. The final week before successful insemination. Available characteristics in the first 6 wk of lactation were: EB, DMI of concentrate and PMR, and plasma NEFA, BHB, and glucose concentration. Available characteristics between calving and successful insemination were: peak production, day of peak production, slope to peak production, slope from peak production until day of successful insemination, and slope in the 3 wk before successful insemination. Peak production was calculated as the greatest 5-day rolling average milk production between calving and successful insemination. Slope to peak production was calculated as the slope in kg milk per day from day 10 in lactation until day of peak production and was computed as peak production minus the 5-day rolling average of the milk production on day 10 in lactation (i.e., the average milk production from day 8 until day 12), divided by the day of peak production minus 10. Slope from peak production until successful insemination was calculated the same way, using the peak production and the 5-day rolling average of the final 5 days before successful insemination. Slope in the last 3 wk before successful insemination was calculated the same way. Available characteristics before successful insemination were: mean milk production, mean fat, protein, and lactose content in the milk, fat to protein ratio, mean BW, mean BCS (final month before successful insemination), mean SCC, and mean plasma insulin and IGF-1 concentration.

2.4 Statistical analysis

Parity (primiparous or multiparous) refers to the parity of the cow after the first calving in the experiment. Statistical analyses were performed by using SAS version 9.4 (SAS Institute Inc., Cary, NC). For all models, plasma NEFA, BHB, and glucose concentration, plasma insulin concentration, SCC, slope to maximum production, and slope from maximum production to successful insemination were transformed to their natural logarithm to approximate a normal distribution. In the model for the 6 wk after calving 2 in the experiment, DMI of concentrate was transformed to the 10th power to approximate a normal distribution. Values are presented as LSM ± SEM. All *P*-values of pair-wise comparisons of LSM were corrected with the Bonferroni-adjustment.

First, data was analyzed separately for 4 periods during different phases of the lactation: first 305 days after calving 1, 8 wk around the end of the VWP (i.e., around the start of the insemination period; d -28 to d 28 relative to the end of the VWP), 8 wk around the successful insemination (i.e., the insemination that was later confirmed to be successful and resulted in calving 2), and the pregnancy period (i.e., period of 280 d before calving 2). Second, data was analyzed from 2 wk before until 6 wk after calving 2. Third, individual cow characteristics as monitored before successful insemination were used to predict lactation performance of cows after different VWP. The first 6 wk after calving 1 in the experiment were not analyzed separately as no effect of VWP can be apparent during that time; this data was only used in the prediction models.

First 305 days after calving 1

During the first 305 days after calving 1, plasma insulin and IGF-1 concentration, BCS, BW, ΔBW, and the ratio of NE-FPCM to NE-BW were analyzed for fixed effects of VWP (VWP50, VWP125, or VWP200), parity (primiparous or multiparous), lactation week (wk 1 – wk 44), and their two-way interactions. A repeated measurements model (PROC MIXED) was used for this analysis, with a repeated effect of lactation week with cow as the repeated subject. Plasma insulin and IGF-1 concentration were measured once every 2 wk between 7 wk and 44 wk in lactation, and were averaged per 2 wk. The analyses were done for all cows (N = 153), and for cows that had a second calf in the experiment (n = 127).

Eight weeks around the end of the VWP

During the 8 wk around the end of the VWP, plasma insulin and IGF-1 concentration, BCS, BW, ΔBW, and the ratio of NE-FPCM to NE-BW were analyzed for fixed effects of VWP (VWP50, VWP125, or VWP200), parity (primiparous or multiparous), week relative to the end of the VWP (-3, -1, 1, 3; where d -28 to -14 is wk -3; d -14 to -1 is wk -1; d 1 to 14 is wk 1; and d 14 to 28 is wk 3), and their two-way interactions. A repeated measurements model (PROC MIXED) was used for this analysis, with a repeated effect of lactation week with cow as the repeated subject. The analyses were done for all cows that reached the end of the VWP (n = 151), and for cows that had a second calf in the experiment (n = 127).

Eight weeks around successful insemination

During the 8 wk around successful insemination, plasma insulin and IGF-1 concentration, BCS, BW, ΔBW, and the ratio of NE-FPCM to NE-BW were analyzed for fixed effects of VWP (VWP50, VWP125, or VWP200), parity (primiparous or multiparous), week relative to successful insemination (d -28 to d 28; wk -3, -1, 1, 3), and their two-way interactions. A repeated measurements model (PROC MIXED) was used for this analysis, with a repeated effect of lactation week with cow as the repeated subject. The analysis was done for all cows that had a successful insemination (n = 127).

Pregnancy period

During the pregnancy period, plasma insulin and IGF-1 concentration were analyzed for fixed effects of VWP, parity (primiparous or multiparous), week relative to calving 2 (wk -40 – wk -1), and their two-way interactions. A repeated measurements model (PROC MIXED) was used for this analysis, with a repeated effect of week to calving with cow as the repeated subject. The analysis was done for all cows that had a second calf in the experiment (n = 127). For the final 34 wk before dry-off (40 wk pregnancy minus 6 wk dry period), BCS, BW, ΔBW, and the ratio of NE-FPCM to NE-BW were analyzed for fixed effects of VWP, parity, week relative to dry-off (wk -34 – wk -1), and their two-way interactions. A repeated measurements model (PROC MIXED) was used for this analysis, with a repeated effect of week to dry-off with cow as the repeated subject. The analysis was done for all cows that had a second calf and a dry period in the experiment (n = 124).

Two weeks before until 6 weeks after calving 2

During the period around calving 2, BCS, BW, DMI, EB, and plasma NEFA, BHB, glucose, insulin and IGF-1 concentration were analyzed for fixed effects of VWP (VWP50, VWP125, or VWP200), parity (primiparous or multiparous, referring to the parity of the cow in the first lactation in the experiment), week relative to calving 2 (wk -2 – wk 6), and their two-way interactions. A repeated measurements model (PROC MIXED) was used for this analysis, with a repeated effect of lactation week with cow as the repeated subject. The periods before calving and after calving were analyzed separately. The analysis was done for all cows that had a second calf in the experiment (n = 127).

Moreover, to evaluate the effect of CInt on the period around calving 2, cows were categorized into 3 groups with different CInt, based on a similar number of cows per group: CInt-1: <415 d (n = 43), CInt-2: 415 until 484 d (n = 44), or CInt-3: ≥485 d (n = 40). A repeated measurements model (PROC MIXED) was used to analyze the effect of CInt group, parity (primiparous or multiparous, referring to the parity of the cow in the first lactation in the experiment), week relative to calving 2 (wk -2 – wk 6), and their two-way interactions on BCS, BW, DMI, EB, and plasma NEFA, BHB, glucose, insulin and IGF-1 concentration. The model had a repeated effect of lactation week with cow as the repeated subject.

Cow characteristics as predictor for lactation performance

Cow characteristics between calving 1 and successful insemination were evaluated as a predictor for lactation performance after different VWP. Evaluations were based on data of cows that had a second calf and a dry period in the experiment (n = 124). Cow characteristics were extracted from prior data, and characteristics in three periods: 1. The first 6 weeks of lactation, 2. Between calving and successful insemination, and 3. The final week before successful insemination. The 3 variables for lactation performance were: FPCM production in the final 6 wk before dry-off, BCS in the final 12 wk before dry-off, and FPCM production per day of CInt. First, the effect of VWP, parity, and their interaction on these variables for lactation performance was tested with a general linear model in SAS (PROC MIXED). Second, the effect of each cow characteristic on these variables for lactation performance was tested in a univariate analysis, but always including the effect of parity, with a general linear model in SAS (PROC MIXED). The VWP was not included in the prediction models, to predict lactation performance based on cow characteristics before successful insemination independent of the VWP. Third, when P-value was < 0.20, the cow characteristic was included in the multivariable model. The multivariable model always included parity as fixed effect. The cow characteristics and their interaction with parity stayed in the model if $P < 0.05$ by using backward selection. Finally, the adjusted R^2 of the final multivariable model was compared with the adjusted R^2 of the model with only VWP, parity, and their interaction when significant, to investigate added value of the cow characteristics in the model instead of VWP to predict the variable for lactation performance. Adjusted R^2 was calculated as follows:

$$\text{Adjusted } \mathrm{R}^2 = 1 - \frac{(1-\mathrm{R}^2)\times(n-1)}{(n-1-k)},$$

where n is the sample size in the analysis, and k is the number of independent variables in the analysis.

3 Results

One cow was culled on day 43 after calving, therefore 153 cows were included in the analyses (41 primiparous and 112 multiparous cows).

3.1 Effect of voluntary waiting period on lactation performance during different phases of the lactation

First 305 days after calving 1

Multiparous cows in VWP200 had a greater plasma insulin concentration compared with multiparous cows in VWP50 ($P < 0.01$) or VWP125 ($P < 0.01$) during the first 305 days after calving 1 (Table 1, Figure 1). Multiparous cows in VWP200 had a greater plasma IGF-1 concentration compared with multiparous cows in VWP125 ($P < 0.01$), and the plasma IGF-1 concentration of multiparous cows in VWP50 did not differ from the other 2 VWP. Multiparous cows in VWP200 had a lower FPCM production ($P = 0.02$) and a lower ratio of NE-FPCM to NE-BW ($P < 0.01$) compared with multiparous cows in VWP125. The FPCM production and the ratio of NE-FPCM to NE-BW of multiparous cows in VWP50 did not differ from the other 2 VWP.

Primiparous cows in VWP125 had a greater plasma insulin concentration compared with primiparous cows in VWP50 ($P = 0.01$) during the first 305 days after calving 1, and the plasma insulin concentration of primiparous cows in VWP200 did not differ from the other 2 VWP. During the first 305 days after calving 1, the VWP did not affect the plasma IGF-1 concentration, the FPCM production, or the ratio of NE-FPCM to NE-BW of primiparous cows. For all cows (i.e., primiparous cows and multiparous cows together), the effect of VWP on the plasma IGF-1 concentration, FPCM production, and ratio of NE-FPCM to NE-BW depended on week in lactation. Moreover, for all cows, the VWP did not affect the BCS, BW, or ΔBW.

When culled and non-pregnant cows were excluded, primiparous cows in VWP200 had a lower plasma IGF-1 concentration (146.4 ng/mL) compared with primiparous cows in VWP50 (178.5 ng/mL; $P = 0.02$) or VWP125 (176.4 ng/mL, $P = 0.03$). In the analysis excluding culled and non-pregnant cows, the FPCM production or the ratio of NE-FPCM to NE-BW did not differ among the different VWP in the two parity classes, but the effect of VWP on the FPCM production or the ratio of NE-FPCM to NE-BW still depended on week in lactation. In all other analyses excluding culled and non-pregnant cows, the results were similar to the analyses including culled and non-pregnant cows.

When considering cows that did not conceive or were culled during the first lactation in the experiment as a separate group (n = 26), the plasma insulin concentration was 14.6 ± 2.4 µU/mL for primiparous cows (n = 5) and 15.7 ± 1.4 µU/mL for multiparous cows (n = 21), and the plasma IGF-1 concentration was 147 ± 16 ng/mL for primiparous cows and 133 ± 11 ng/mL for multiparous cows. For this group, the BCS was 2.4 ± 0.1 for primiparous cows and 2.4 ± 0.1 for multiparous cows, the BW was 555 ± 11 kg for primiparous cows and 692 ± 17 kg for multiparous cows, and the ΔBW was 0.70 ± 1.07 kg/wk for primiparous cows and -0.74 ± 0.88 kg/wk for multiparous cows. The FPCM production was 26.0 ± 1.2 kg/d for primiparous cows and 34.9 ± 1.3 kg/d for multiparous cows, and the ratio of NE-FPCM to NE-BW was 2.4 ± 0.1 for primiparous cows and 2.7 ± 0.1 for multiparous cows.

Eight weeks around the end of the VWP

Multiparous cows in VWP200 had a greater plasma insulin and IGF-1 concentration compared with multiparous cows in VWP125 ($P < 0.05$) or VWP50 ($P < 0.01$) during the 8 wk around the end of the VWP (i.e., around the start of the insemination period) (Figure 2). For multiparous cows, the FPCM production and the ratio of NE-FPCM to NE-BW was lowest in VWP200, intermediate in VWP125, and greatest in VWP50 ($P < 0.01$ for all comparisons).

For primiparous cows, the VWP did not affect the plasma insulin and IGF-1 concentration, the FPCM production, or the ratio of NE-FPCM to NE-BW during the 8 wk around the end of the VWP. For both primiparous and multiparous cows, the effect of VWP on the FPCM production and the ratio of NE-FPCM to NE-BW depended on week around the end of the VWP. The BW or ΔBW did not differ among the 3 VWP in the different weeks or months around the end of the VWP. Moreover, for both primiparous and multiparous cows, VWP did not affect the BCS.

When culled and non-pregnant cows were excluded, the effect of VWP on the plasma insulin concentration did not depend on parity, and all cows in VWP200 had a greater plasma insulin concentration (16.3 µU/mL) compared with all cows in VWP50 (13.0 µU/mL, $P = 0.04$). Moreover, all cows in VWP200 tended to have a greater BW compared with all cows in VWP50 (650 vs. 619 kg, $P = 0.06$). In all other analyses excluding culled and non-pregnant cows, the results were similar to the analyses including culled and non-pregnant cows.

Table 1. Plasma insulin and IGF-1 concentration, body condition, fat- and protein-corrected milk, and energy partitioning for different periods in the lactation of primiparous and multiparous cows with a voluntary waiting period after calving until first insemination of 50, 125, or 200 days (VWP50, VWP125, VWP200) (LSM ± maximum SEM or CI)

	Primiparous cows				Multiparous cows				*P*-value[1]					
	VWP50	VWP125	VWP200	SEM (CI)	VWP50	VWP125	VWP200	SEM (CI)	VWP	P	W	VWP×P	VWP×W	P×W
First 305 days after calving 1– n cows	14	15	12		39	34	39							
Insulin (µU/mL)[2]	11.0^{b}	13.4^{a}	12.6^{ab}	(10.1-14.6)	12.3^{b}	12.5^{b}	14.5^{a}	(11.7-15.3)	<0.01	0.05	<0.01	<0.01	0.07	0.41
IGF-1[3] (ng/mL)	174.1	174.2	151.5	7.2	138.9^{ab}	126.4^{b}	145.5^{a}	4.3	0.34	<0.01	<0.01	<0.01	0.02	0.51
BCS (first 11 mo)	2.6	2.6	2.4	0.08	2.3	2.3	2.4	0.05	0.38	<0.01	0.08	0.19	0.35	0.13
BW (kg)	593	578	592	15	700	692	687	9	0.60	<0.01	<0.01	0.73	0.57	0.50
ΔBW[4] (kg/wk)	2.4	1.8	1.9	0.4	1.3	0.8	1.1	0.2	0.19	<0.01	<0.01	0.81	0.46	0.03
FPCM[5] (kg/d)	26.6	27.7	28.0	1.1	34.3^{ab}	34.8^{a}	32.3^{b}	0.6	0.39	<0.01	<0.01	0.09	<0.01	<0.01
Ratio NE-FPCM : NE-BW[6]	2.3	2.4	2.4	0.07	2.6^{ab}	2.7^{a}	2.5^{b}	0.04	0.18	<0.01	<0.01	0.06	<0.01	<0.01
8 wk around end VWP – n cows	14	15	12		39	34	37							
Insulin (µU/mL)[2]	12.6	16.5	14.3	(10.4-19.8)	13.4^{b}	13.7^{b}	17.6^{a}	(11.9-19.8)	0.04	0.68	0.42	0.05	0.70	0.81
IGF-1 (ng/mL)	163.3	182.0	174.5	11.7	115.2^{b}	126.4^{b}	174.5^{a}	7.0	<0.01	<0.01	0.28	<0.01	0.54	0.80
BCS (2 mo around end VWP)	2.7	2.6	2.4	0.13	2.4	2.3	2.5	0.08	0.83	0.04	0.37	0.19	0.91	0.74
BW (kg)	561	571	595	16	683	685	695	9.5	0.17	<0.01	0.16	0.67	0.04	0.03
ΔBW (kg/wk)	2.2	2.1	1.7	1.1	-1.3	0.014	0.72	0.70	0.67	<0.01	0.58	0.35	0.05	0.68
FPCM (kg/d)	29.9	28.0	28.0	1.4	42.4^{a}	36.9^{b}	30.1^{c}	0.8	<0.01	<0.01	0.01	<0.01	<0.01	0.13
Ratio NE-FPCM : NE-BW	2.7	2.5	2.4	0.10	3.3^{a}	2.9^{b}	2.3^{c}	0.06	<0.01	<0.01	<0.01	<0.01	<0.01	0.32

[a-c] Different superscript indicates a difference among LSM within the row and within the parity ($P < 0.05$).

[1] VWP = voluntary waiting period. P = parity (primiparous or multiparous) in the first lactation in the experiment. W = week in lactation; month for BCS.

[2] Transformed data are back transformed, and confidence interval is shown.

[3] IGF-1 = insulin-like growth factor 1.

[4] ΔBW = difference between mean BW in one week and mean BW in the next week.

[5] FPCM = fat- and protein-corrected milk.

[6] Ratio NE-FPCM : NE-BW = ratio between the net energy (kJ) for FPCM and net energy for maintenance.

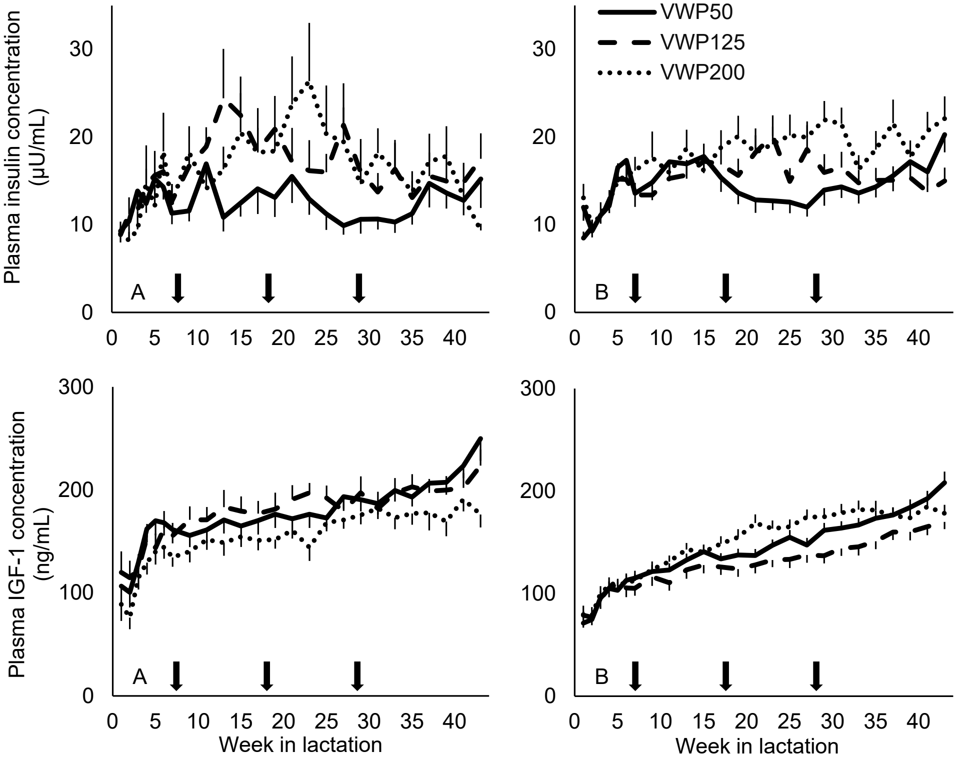


Figure 1. Plasma insulin concentration (µU/mL) and plasma IGF-1 concentration (ng/mL) for the first 305 days after calving 1 of primiparous (A) and multiparous (B) cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, VWP200). Arrows depict the week in lactation from which insemination started for the respective VWP groups. Error bars are depicted either below or above the graphs.

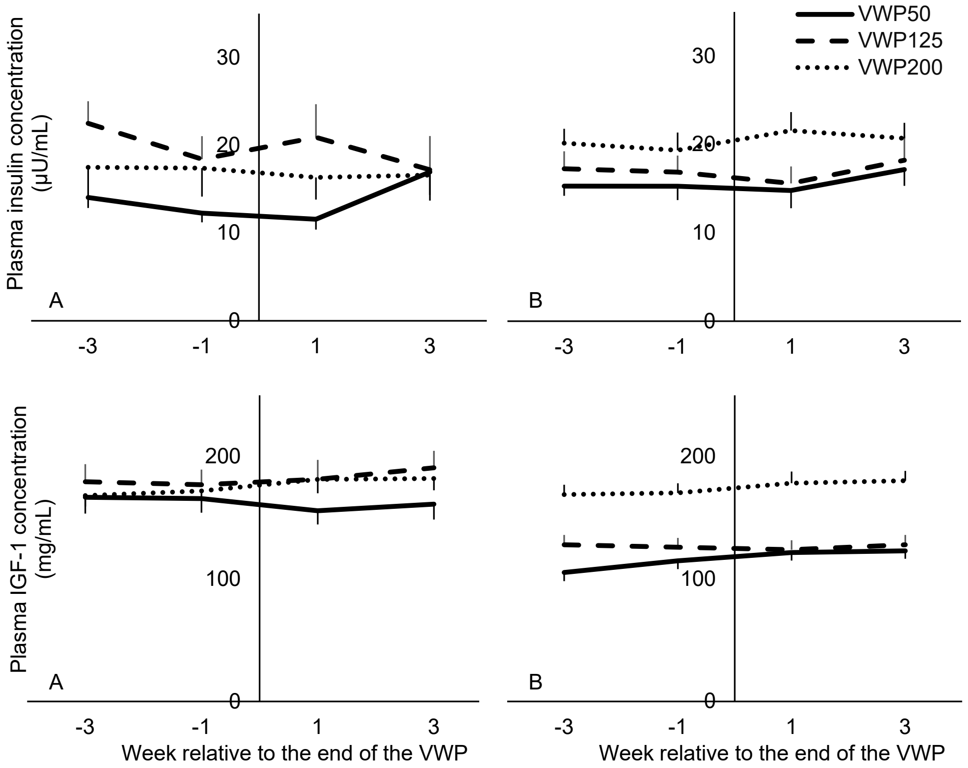


Figure 2. Plasma insulin concentration (µU/mL) and plasma IGF-1 concentration (ng/mL) for the 8 wk around the end of the VWP of primiparous (A) and multiparous (B) cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, VWP200). Error bars are depicted either below or above the graphs.

Eight weeks around successful insemination

All cows in VWP200 tended to have a greater plasma insulin concentration compared with all cows in VWP50 (15.6 vs. 12.8 µU/mL, *P* = 0.09), and the plasma insulin concentration of all cows in VWP125 did not differ from the other 2 VWP during the 8 wk around successful insemination. Multiparous cows in VWP200 had a greater plasma IGF-1 concentration compared with multiparous cows in VWP125 (*P* = 0.01) or VWP50 (*P* < 0.01) (Table 2). For multiparous cows, the FPCM production and the ratio of NE-FPCM to NE-BW was lowest in VWP200, intermediate in VWP125, and greatest in VWP50 (P ≤ 0.01 for all comparisons).

For primiparous cows, the VWP did not affect the plasma IGF-1 concentration, FPCM production, and ratio of NE-FPCM to NE-BW during the 8 wk around successful insemination. For all cows, the VWP did not affect the BCS, BW, or ΔBW.

Pregnancy period

For multiparous cows, the plasma insulin concentration was greatest in VWP200, intermediate in VWP125, and lowest in VWP50 (P ≤ 0.02 for all comparisons). Multiparous cows in VWP200 had a greater plasma IGF-1 concentration compared with multiparous cows in VWP125 (*P* = 0.04) or VWP50 (*P* < 0.01).

Primiparous cows in VWP125 had a greater plasma insulin concentration compared with primiparous cows in VWP50 (*P* < 0.01), and the plasma insulin concentration of primiparous cows in VWP200 did not differ from the other 2 VWP (Figure 3) during the pregnancy period. Primiparous cows in VWP125 tended to have a greater plasma IGF-1 concentration compared with primiparous cows in VWP200, and the plasma IGF-1 concentration of primiparous cows in VWP50 did not differ from the other 2 VWP.

During the final 34 wk before dry-off (40 wk – 6 wk dry period), all cows in VWP200 had greater BW (697 kg) compared with all cows in VWP50 (665 kg; *P* = 0.03). The BW of all cows in VWP125 (676 kg) did not differ from the other 2 VWP. Multiparous cows in VWP200 had greater BCS compared with multiparous cows in VWP125 or VWP50 (*P* < 0.01) and gained more BW per week compared with multiparous cows in VWP50 (*P* < 0.01). The ΔBW of multiparous cows in VWP125 did not differ from the other 2 VWP. During these final 34 wk before dry-off, for multiparous cows, the FPCM production and the ratio of NE-FPCM to NE-BW was lowest in VWP200, intermediate in VWP125, and greatest in VWP50 (P ≤ 0.01 for all comparisons). During the final 34 wk before dry-off, the VWP did not affect the BCS or ΔBW of primiparous cows. For all cows, the effect of VWP on the plasma IGF-1 concentration, FPCM production, ratio of NE-FPCM to NE-BW, and BCS depended on week or month before calving or dry-off.

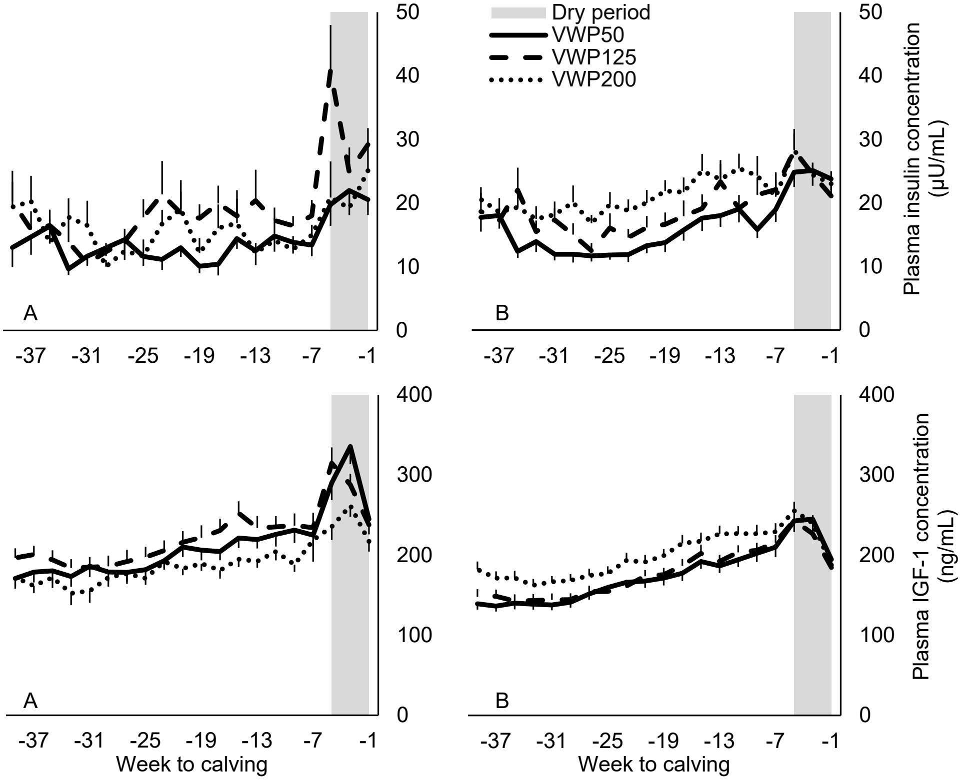


Figure 3. Plasma insulin concentration (μU/mL) and plasma IGF-1 concentration (ng/mL) for the pregnancy period of primiparous (A) and multiparous (B) cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, VWP200). The dry period of the cows is illustrated in gray. Error bars are depicted either below or above the graphs.

Table 2. Plasma insulin and IGF-1 concentration, body condition, fat- and protein-corrected milk, and energy partitioning for different periods in the lactation of primiparous and multiparous cows with a voluntary waiting period after calving until first insemination of 50, 125, or 200 days (VWP50, VWP125, VWP200) (LSM ± maximum SEM or CI)

	Primiparous cows				Multiparous cows				*P*-value[1]					
	VWP50	VWP125	VWP200	SEM (CI)	VWP50	VWP125	VWP200	SEM (CI)	VWP	P	W	VWP×P	VWP×W	P×W
n cows	13	14	9		34	28	29							
8 wk around successful insemination														
Insulin (µU/mL)[2]	11.9	15.4	15.2	(9.8-19.2)	13.9	15.1	15.9	(12.3-18.2)	0.047	0.41	0.73	0.55	0.72	0.80
IGF-1[3] (ng/mL)	176.3	198.9	166.7	13.5	135.2^{b}	145.2^{b}	178.8^{a}	7.7	0.14	<0.01	0.34	<0.01	0.46	0.78
BCS	2.6	2.6	2.4	0.15	2.2	2.3	2.5	0.09	0.81	0.06	0.62	0.12	0.28	0.58
BW (kg)	576	588	604	17	680	687	703	9.8	0.15	<0.01	<0.01	0.97	0.41	0.22
ΔBW[4] (kg/wk)	3.2	3.1	2.3	1.0	1.2	1.5	1.7	0.6	0.93	0.03	<0.01	0.66	0.11	0.18
FPCM[5] (kg/d)	29.3	27.7	28.8	1.7	38.1^{a}	34.0^{b}	28.7^{c}	1.0	<0.01	<0.01	<0.01	<0.01	0.62	<0.01
Ratio NE-FPCM : NE-BW[6]	2.6	2.4	2.5	0.13	3.0^{a}	2.6^{b}	2.2^{c}	0.07	<0.01	0.15	<0.01	<0.01	0.71	<0.01
Pregnancy period														
Insulin (µU/mL)[2]	11.9^{b}	15.4^{a}	13.6^{ab}	(10.8-17.0)	13.9^{c}	15.8^{b}	18.5^{a}	(13.0-19.7)	<0.01	<0.01	<0.01	<0.01	0.42	0.52
IGF-1 (ng/mL)	210.8	221.7†	191.5†	9.3	175.3^{b}	178.2^{b}	198.5^{a}	5.3	0.56	<0.01	<0.01	<0.01	<0.01	<0.01
BCS[7]	2.6	2.7	2.5	0.13	2.4^{b}	2.6^{b}	3.0^{a}	0.07	0.04	0.38	<0.01	<0.01	<0.01	<0.01
BW (kg)[7]	620	630	648	17	709	722	747	9.6	0.04	<0.01	<0.01	0.91	0.33	0.96
ΔBW (kg/wk)[7]	2.9	3.4	2.7	0.5	2.5^{b}	2.9^{ab}	3.6^{a}	0.2	0.20	0.93	<0.01	0.07	0.72	0.31
FPCM (kg/d)[7]	26.8	25.7	26.9	1.5	30.0^{a}	26.6^{b}	22.6^{c}	0.8	<0.01	0.92	<0.01	<0.01	0.02	<0.01
Ratio NE-FPCM : NE-BW[7]	2.3	2.1	2.2	0.11	2.3^{a}	2.0^{b}	1.7^{c}	0.06	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01

$^{a\text{-}c}$Different superscript indicates a difference among LSM within the row and within the parity ($P < 0.05$).

[1] VWP = voluntary waiting period. P = parity (primiparous or multiparous) in the first lactation in the experiment. W = week in lactation; month for BCS.

[2] Transformed data are back transformed, and confidence interval is shown.

[3] IGF1 = insulin-like growth factor 1.

[4] ΔBW = difference between mean BW in one week and mean BW in the next week.

[5] FPCM = fat- and protein-corrected milk.

[6] Ratio NE-FPCM : NE-BW = ratio between the net energy (kJ) for FPCM and net energy for maintenance.

[7] Analyzed in the final 34 weeks before dry-off, only cows with dry period (n = 124).

†Similar symbol indicates a trend in difference among LSM within the row and within the parity ($P < 0.10$).

3.2 Effect of voluntary waiting period on metabolic status around calving 2

From 2 weeks before until 6 weeks after calving 2 per voluntary waiting period group

Multiparous cows in VWP200 had a lower plasma BHB concentration compared with multiparous cows in VWP50 ($P = 0.02$), and the plasma BHB concentration of multiparous cows in VWP125 did not differ from the other 2 VWP (Table 3) during the 2 wk before calving 2. The VWP did not affect the plasma BHB concentration of primiparous cows before calving 2. For all cows, the VWP did not affect the BW, DMI, EB, or the plasma NEFA, glucose, insulin or IGF-1 concentration during the 2 wk before this calving (Figure 4).

During the 6 wk after calving 2, for multiparous cows, BCS was greatest in VWP200, intermediate in VWP125, and lowest in VWP50. Multiparous cows in VWP200 had greater plasma NEFA concentration compared with multiparous cows in VWP50 ($P < 0.01$) or VWP125 ($P = 0.04$). In wk 1 after calving 2, all cows in VWP200 had a more negative EB compared with all cows in VWP50 (-302 vs. -160 kJ/BW0.75, $P = 0.02$). Moreover, all cows in VWP125 had a greater plasma BHB concentration (0.80 mmol/L) compared with all cows in VWP50 (0.65 mmol/L, $P < 0.01$), and the plasma BHB concentration of all cows in VWP200 (0.76 mmol/L) did not differ from the other 2 VWP after calving 2.

The VWP did not affect the BCS or plasma NEFA concentration of primiparous cows. For all cows, the VWP did not affect the BW, DMI, or the plasma glucose, insulin or IGF-1 concentration during the 6 wk after calving 2.

Table 3. Body condition, energy balance and plasma metabolites and hormones from 2 wk before until 6 wk after calving 2 of primiparous and multiparous cows with a voluntary waiting period after calving until first insemination of 50, 125, or 200 days (VWP50, VWP125, VWP200) (LSM±SEM or CI)

	Primiparous cows				Multiparous cows				*P*-value[1]					
	VWP50	VWP125	VWP200	SEM (CI)	VWP50	VWP125	VWP200	SEM (CI)	VWP	P	W	VWP×P	VWP×W	P×W
n cows	13	14	9		34	28	29							
Wk -2 – wk -1														
BW (kg)	718	708	749	42	768	773	752	21	0.94	0.08	0.36	0.56	0.66	0.96
DMI[2] concentrate (kg/d)	1.8	1.7	1.7	0.07	1.7	1.7	1.6	0.04	0.27	0.11	<0.01	0.57	0.88	0.96
DMI PMR[3] (kg/d)	14.0	13.9	14.8	0.81	14.6	13.7	13.6	0.44	0.57	0.60	<0.01	0.35	0.76	0.05
EB[4] (kJ/BW$^{0.75}$)	246	250	261	47	232	221	206	26	0.98	0.23	0.26	0.82	0.61	0.9999
NEFA[5] (mmol/L)[6]	0.09	0.09	0.06	(0.04-0.12)	0.09	0.11	0.11	(0.07-0.14)	0.51	0.05	<0.01	0.20	0.81	0.20
BHB[7] (mmol/L)[6]	0.50	0.57	0.55	(0.44-0.65)	0.49^{a}	0.46^{ab}	0.41^{b}	(0.38-0.52)	0.33	<0.01	0.60	0.03	0.41	0.42
Glucose (mmol/L)[6]	3.46	3.57	3.61	(3.36-3.74)	3.53	3.48	3.51	(3.41-3.60)	0.41	0.31	0.18	0.12	0.08	0.18
Insulin (μU/mL)[6]	18.5	26.3	22.2	(14.5-33.3)	21.9	18.7	20.9	(15.9-25.3)	0.60	0.39	<0.01	0.05	0.56	0.51
IGF-1[8] (ng/mL)	248.2	240.2	215.0	18.4	199.3	185.2	191.9	10.5	0.34	<0.01	<0.01	0.50	0.18	0.56
Wk 1 – wk 6														
BCS	2.5	2.9	2.8	0.2	2.7^{b}	3.0^{b}	3.5^{a}	0.1	<0.01	0.01	0.03	0.08	0.58	0.96
BW (kg)	637	630	647	20	702	694	725	12	0.30	<0.01	0.09	0.89	0.98	0.96
DMI concentrate (kg/d)[6]	8.0	8.0	7.8	(7.4-8.2)	7.9	7.8	7.8	(7.6-8.0)	0.34	0.18	<0.01	0.44	0.39	<0.01
DMI PMR (kg/d)	15.1	14.3	14.8	0.7	15.8	14.4	13.9	0.4	0.05	0.97	<0.01	0.37	0.20	<0.01
EB (kJ/BW$^{0.75}$)	-202	-188	-132	51	-203	-213	-296	31	0.93	0.046	<0.01	0.08	<0.01	0.61
NEFA (mmol/L)[6]	0.21	0.23	0.19	(0.14-0.29)	0.26^{b}	0.30^{b}	0.41^{a}	(0.22-0.48)	0.21	<0.01	<0.01	0.01	0.19	0.98
BHB (mmol/L)[6]	0.65	0.80	0.69	(0.55-0.93)	0.66	0.81	0.84	(0.60-0.93)	<0.01	0.21	<0.01	0.37	0.26	0.21
Glucose (mmol/L)[6]	3.14	3.07	3.18	(2.91-3.39)	3.16	3.07	3.04	(2.93-3.27)	0.51	0.54	<0.01	0.51	0.89	0.76
Insulin (μU/mL)[6]	10.0	12.2	11.4	(8.4-14.4)	11.1	10.3	9.3	(8.3-12.3)	0.53	0.17	<0.01	0.08	0.32	0.25
IGF-1 (ng/mL)	109.3	97.6	90.8	12.2	75.8	77.5	67.7	7.3	0.36	<0.01	<0.01	0.71	0.37	0.65

[a,b] Different superscript indicates a difference among LSM within the row and within the parity ($P < 0.05$).

[1] VWP = voluntary waiting period. P = parity (primiparous or multiparous) in the first lactation in the experiment. W = week relative to calving; month for BCS.

[2] DMI = dry matter intake.

[3] PMR = partially mixed ration, consisting of grass silage, corn silage, soybean meal, and wheat meal.

[4] EB = energy balance, calculated as the difference between intake of NE and requirements of NE for maintenance, milk production, and pregnancy.

[5] NEFA = non-esterified fatty acids.

[6] Transformed data are back transformed, and confidence interval is shown.

[7] BHB = β-hydroxybutyrate.

[8] IGF1 = insulin-like growth factor 1.

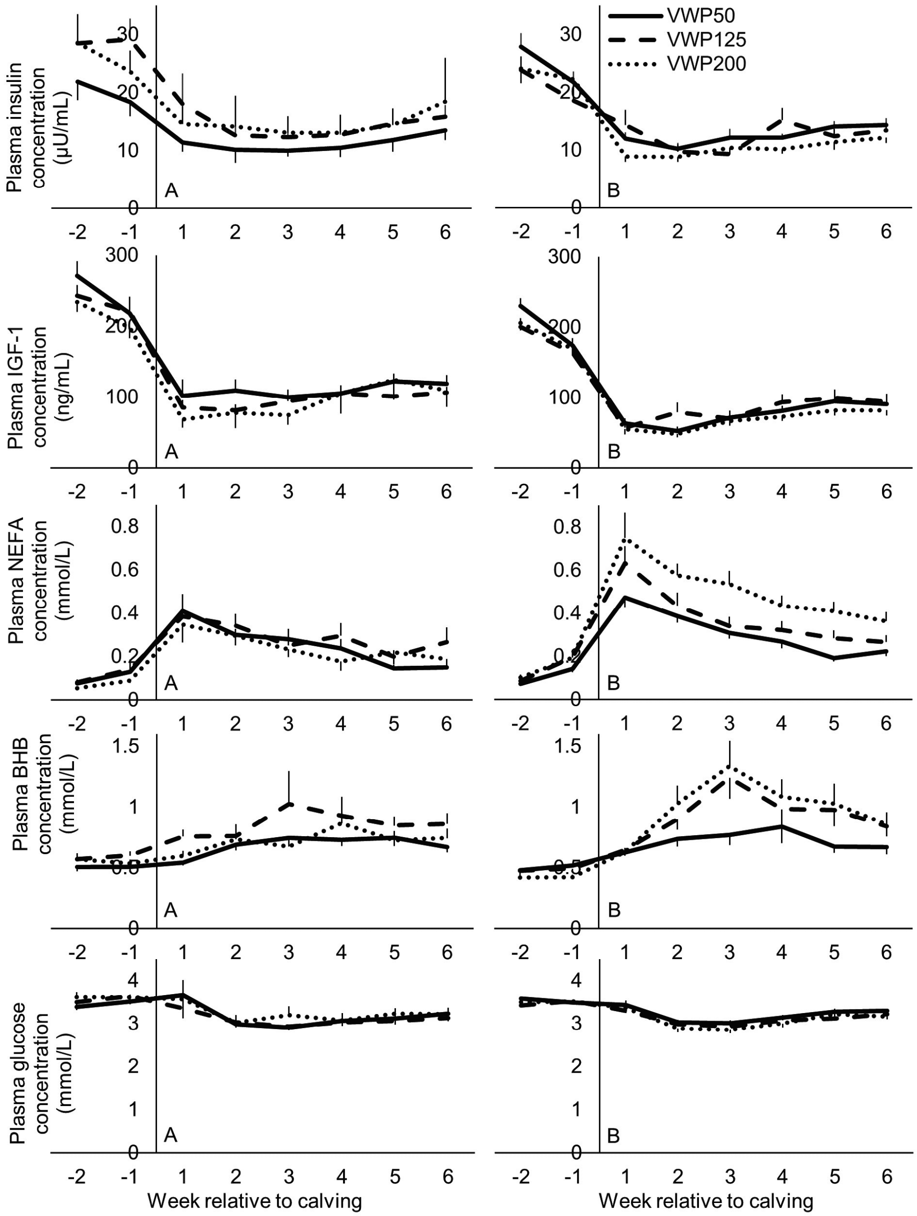


Figure 4. Plasma insulin (µU/mL), IGF-1 (ng/mL), NEFA (mmol/L), BHB (mmol/L), and glucose (mmol/L) concentration from 2 wk before until 6 wk after calving 2 of primiparous (A) and multiparous (B) cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, VWP200). Parity (primiparous or multiparous cows) refers to the parity of the cow during the first lactation in the experiment. Error bars are depicted either below or above the graphs.

From 2 weeks before until 6 weeks after calving 2 per calving interval group

During the 2 wk before calving 2, the CInt did not affect the BW, DMI, EB, or the plasma NEFA, BHB, glucose, insulin or IGF-1 concentration (Table 4). Multiparous cows in CInt-1 had a greater plasma insulin concentration compared with multiparous cows in CInt-2 (*P* = 0.02), and the plasma insulin concentration of multiparous cows in CInt-3 did not differ from the other 2 CInt during the 6 wk after calving 2. Multiparous cows in CInt-3 had a more negative EB compared with multiparous cows in CInt-1, and the EB of multiparous cows in CInt-2 did not differ from the other 2 CInt. For all cows, the effect of CInt on the BCS, DMI, and EB during the 6 wk after calving 2 depended on month or week after calving. In month 1 after calving, BCS was lower for all cows in CInt-1 (2.5) compared with all cows in CInt-2 (3.0; *P* < 0.01) or CInt-3 (3.4; *P* < 0.01). In week 1 after calving, the DMI from PMR was higher for all cows in CInt-1 (14.3 kg/d) compared with all cows in CInt-2 (12.0 kg/d, *P* < 0.01) or CInt-3 (11.6 kg/d, *P* < 0.01). In week 6 after calving, the DMI from concentrate was higher for all cows in CInt-1 (8.8 kg/d) and CInt-2 (8.8 kg/d) compared with all cows in CInt-3 (8.6 kg/d, *P* < 0.01). In week 1 after calving, the EB was less negative for all cows in CInt-1 (-131 kJ/BW0.75) compared with all cows in CInt-2 (-261 kJ/BW0.75, *P* = 0.03) or CInt-3 (-293 kJ/BW0.75, *P* < 0.01). During the 6 wk after calving 2, all cows in CInt-1 had lower plasma NEFA and BHB concentration (NEFA: 0.21 mmol/L; BHB: 0.64 mmol/L) compared with all cows in CInt-2 (NEFA: 0.28 mmol/L; BHB: 0.82 mmol/L; *P* < 0.01) or CInt-3 (NEFA: 0.30 mmol/L, *P* < 0.01; BHB: 0.76 mmol/L, *P* = 0.04). During the 6 wk after calving 2, the VWP did not affect the plasma insulin concentration of primiparous cows. For all cows, the VWP did not affect the BW, or the plasma glucose or IGF-1 concentration.

Table 4. Body condition, energy balance and plasma metabolites and hormones from 2 wk before until 6 wk after calving 2 of primiparous and multiparous cows with a voluntary waiting period after calving until first insemination of 50, 125, or 200 days (VWP50, VWP125, VWP200) per calving interval (CInt) group (CInt-1: <415 d; CInt-2: 415-484 d; CInt-3: ≥485 d) (LSM±SEM or CI)

	Primiparous cows				Multiparous cows				*P*-value^1					
	CInt-1	CInt-2	CInt-3	SEM (CI)	CInt-1	CInt-2	CInt-3	SEM (CI)	CInt	P	W	CInt×P	CInt×W	P×W
n cows	12	13	11		31	31	29							
Wk -2 – wk -1														
BW (kg)	705	718	742	37	756	765	776	22	0.58	0.04	0.32	0.96	0.44	0.90
DMI^2 concentrate (kg/d)	1.8	1.7	1.7	0.07	1.7	1.7	1.7	0.04	0.59	0.10	<0.01	0.69	0.98	0.98
DMI PMR^3 (kg/d)	14.2	13.8	14.4	0.7	14.6	13.6	13.7	0.4	0.45	0.77	<0.01	0.63	0.63	0.048
EB^4 (kJ/BW$^{0.75}$)	252	267	226	40	253	219	184	26	0.30	0.24	0.23	0.66	0.13	0.92
NEFA^5 (mmol/L)^6	0.08	0.09	0.07	(0.05-0.13)	0.08	0.10	0.12	(0.07-0.14)	0.45	0.08	<0.01	0.18	0.30	0.18
BHB^7 (mmol/L)^6	0.51	0.58	0.54	(0.45-0.65)	0.48	0.47	0.43	(0.40-0.52)	0.35	<0.01	0.56	0.19	0.73	0.41
Glucose (mmol/L)^6	3.46	3.52	3.63	(3.35-3.75)	3.49	3.51	3.52	(3.42-3.59)	0.14	0.45	0.18	0.28	0.37	0.19
Insulin (μU/mL)^6	19.4	21.1	26.7	(15.0-34.6)	20.7	19.4	21.8	(16.5-25.6)	0.16	0.39	<0.01	0.45	0.58	0.44
IGF-1^8 (ng/mL)^6	236	251	223	(189-282)	198	193	187	(166-218)	0.44	<0.01	<0.01	0.65	0.09	0.52
Wk 1 – wk 6														
BCS	2.4	2.9	2.9	0.2	2.6	3.1	3.4	0.1	<0.01	<0.01	0.01	0.30	<0.01	0.89
BW (kg)	632	632	647	18	697	692	732	11	0.13	<0.01	0.11	0.66	0.99	0.97
DMI concentrate (kg/d)^6	8.0	8.0	7.8	(7.5-8.2)	7.8	7.9	7.8	(7.6-8.0)	0.22	0.13	<0.01	0.82	0.01	<0.01
DMI PMR (kg/d)	15.3	13.9	15.1	0.6	16.1	14.4	13.9	0.4	<0.01	0.99	<0.01	0.13	<0.01	<0.01
EB (kJ/BW$^{0.75}$)	-179	-209	-144	46	-173^a	-248^{ab}	-293^b	29	0.32	0.049	<0.01	0.097	<0.01	0.58
NEFA (mmol/L)^6	0.18	0.24	0.22	(0.14-0.31)	0.23	0.33	0.41	(0.20-0.48)	<0.01	<0.01	<0.01	0.10	0.43	0.99
BHB (mmol/L)^6	0.62	0.81	0.71	(0.53-0.95)	0.65	0.84	0.80	(0.58-0.93)	<0.01	0.26	<0.01	0.77	0.12	0.23
Glucose (mmol/L)^6	3.13	3.10	3.13	(2.94-3.32)	3.19	2.99	3.09	(2.88-3.31)	0.28	0.62	<0.01	0.46	0.94	0.77
Insulin (μU/mL)^6	10.0	12.3	11.1	(8.4-14.6)	11.5^a	9.0^b	10.4^{ab}	(8.0-12.8)	0.94	0.19	<0.01	<0.01	0.70	0.31
IGF-1 (ng/mL)	104	109	85	11	81	68	71	7	0.27	<0.01	<0.01	0.30	0.66	0.51

[a,b] Different superscript indicates a difference among LSM within the row and within the parity ($P < 0.05$).

[1] CInt = calving interval. P = parity (primiparous or multiparous) in the first lactation in the experiment. W = week relative to calving; month for BCS.

[2] DMI = dry matter intake.

[3] PMR = partially mixed ration, consisting of grass silage, corn silage, soybean meal, and wheat meal.

[4] EB = energy balance, calculated as the difference between intake of NE and requirements of NE for maintenance, milk production, and pregnancy.

[5] NEFA = non-esterified fatty acids.

[6] Transformed data are back transformed, and confidence interval is shown.

[7] BHB = β-hydroxybutyrate.

[8] Insulin-like growth factor 1.

3.3 Cow characteristics as predictor for lactation performance

First, the effects of VWP, parity, and their interaction on the 3 variables for lactation performance and on the 23 cow characteristics were tested (Table 5). Parity affected all variables for lactation performance, as well as part of the cow characteristics in the first 6 weeks of lactation (EB, DMI, plasma glucose concentration), all cow characteristics between calving and pregnancy, and part of the characteristics in the final week before pregnancy (milk production, lactose content, BW, SCC, and plasma IGF-1 concentration). The VWP affected all variables for lactation performance, and part of the cow characteristics, mainly in the final week before pregnancy.

Table 5. Variables for lactation performance and cow characteristics between calving and successful insemination of cows with a voluntary waiting period after calving until first insemination of 50, 125, or 200 days (VWP50, VWP125, VWP200) that completed the first lactation in the experiment and had a dry period (n = 124) (LSM±SEM or CI)

	Voluntary waiting period				*P*-value[1]		
	VWP50	VWP125	VWP200	SEM (CI)	VWP	Par	VWP×Par
Variables for lactation performance							
FPCM[2] end lactation (kg/d)	22.5^{a}	19.4^{b}	18.2^{b}	1.0	<0.01	<0.01	ns
BCS end lactation	2.6^{a}	2.9^{b}	3.2^{b}	0.11	<0.01	<0.01	0.01
FPCM / day CInt[3] (kg/d)	$27.4^{\dagger}$	26.7	$25.2^{\dagger}$	0.73	0.06	<0.01	ns
Cow characteristics							
First 6 wk							
Energy balance (kJ/$BW^{0.75}$)	-210	-203	-191	24.6	0.83	<0.01	ns
DMI[4] concentrate (kg/d)	5.6	5.6	5.5	0.03	0.08	<0.01	ns
DMI PMR[5] (kg/d)	12.1	12.0	11.7	0.29	0.62	<0.01	ns
NEFA[6] (mmol/L)[7]	0.24	0.27	0.26	(0.20-0.31)	0.47	0.37	ns
BHB[8] (mmol/L)[7]	0.69	0.74	0.69	(0.63-0.80)	0.45	0.66	ns
Glucose (mmol/L)[7]	3.35	3.30	3.38	(3.21-3.48)	0.57	<0.01	ns
Between calving and pregnancy							
Peak production (kg milk/d)	39.6	38.9	38.1	1.1	0.58	<0.01	ns
Day of peak production	48	55	52	3.5	0.3	<0.01	ns
Slope to peak (kg milk/d)[7,9]	0.25	0.20	0.23	(0.16-0.29)	0.17	<0.01	ns
Slope peak – pregnancy (kg milk/d)[7]	-0.12^{b}	-0.07^{a}	-0.06^{a}	(0.05-0.14)	<0.01	<0.01	<0.01
Slope final 3 wk to pregnancy (kg milk/d)	-0.05	-0.07	-0.09	0.02	0.21	<0.01	ns
Final week before pregnancy							
Milk production (kg/d)	33.6^{a}	29.4^{b}	26.9^{b}	1.3	<0.01	<0.01	<0.01
Fat %	3.86^{b}	4.25^{a}	4.51^{a}	0.12	<0.01	0.79	ns
Protein %	$3.49^{b\dagger}$	$3.63^{ab\dagger}$	3.76^{a}	0.05	<0.01	0.28	ns
Lactose %	4.60^{a}	4.55^{ab}	4.49^{b}	0.03	0.01	<0.01	0.099
Fat : protein ratio	1.11^{b}	1.17^{ab}	1.20^{a}	0.03	0.02	0.3	ns
BW (kg)	$627^{\dagger}$	639	$653^{\dagger}$	9.2	0.09	<0.01	ns
BCS[10]	2.4	2.4	2.6	0.10	0.18	0.18	ns
SCC (× 1000)[7]	$55^{\dagger}$	76	$93^{\dagger}$	(39-134)	0.08	<0.01	ns
Insulin (μU/mL)[7]	12.8	16.2	14.8	(10.6-19.8)	0.21	0.6	ns
IGF-1[11] (ng/mL)	153.9	169.4	172.8	9.4	0.22	<0.01	0.02
Prior data							
Previous 305-d production (kg)[12]	8,189	8,231	8,147	251	0.97	<0.01	ns
Breeding value persistency	102.4	103.2	102.1	0.7	0.5	0.03	ns

[a,b] Different superscript letters indicate a difference among LSM within the row ($P < 0.05$).

[1] VWP = voluntary waiting period. Par = parity (primiparous or multiparous) in the first lactation in the experiment. ns = non-significant.

[2] FPCM = fat- and protein-corrected milk.

[3] CInt = calving interval.

[4] DMI = dry matter intake.

[5] PMR = partially mixed ration.

[6] NEFA = non-esterified fatty acids.

[7] Transformed data are back transformed, and confidence interval is shown.

[8] BHB = β-hydroxybutyrate.

[9] Slope from day 10 in lactation until day of maximum production.

[10] Average in the final month before pregnancy.

[11] Insulin-like growth factor 1.

[12] Previous (multiparous) or expected (primiparous) 305-d milk production.

[†] Similar symbol indicates a trend in difference among LSM within the row and within the parity ($P < 0.10$).

For the prediction of mean FPCM production in the final 6 wk before dry-off, 9 cow characteristics were selected at $P < 0.20$ from the univariate analyses (Table A1). These were included in the first multivariable model, next to parity and their interactions with parity. After backward selection, the final multivariable model included 3 cow characteristics (Table 6, figure 5A). With this model, the FPCM production end lactation was predicted for primiparous cows and multiparous cows separately, as follows:

Primiparous cows:

$$\mathrm{FPCM}_{end\ lac} = -21.6 + 0.40 \times \mathrm{MP} + 0.32 \times \mathrm{BV}_{pers},$$

Multiparous cows:

$$\mathrm{FPCM}_{end\ lac} = -27.2 + 0.40 \times \mathrm{MP} + 0.32 \times \mathrm{BV}_{pers},$$

where MP is the average milk production in the final week before successful insemination, and BV_{pers} is the breeding value for persistency. The residuals in this model were on average 3.5 kg FPCM/d. The adjusted R^2 of the final multivariable model was 0.43, and the adjusted R^2 of the model with VWP and parity (i.e., the model used in table 5) was 0.19.

Table 6. Final multivariable model[1] for prediction of fat- and protein-corrected milk (FPCM) end lactation for cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, VWP200) that completed the first lactation in the experiment and had a dry period (n = 124) (LSM±SEM or β±SE)

Cow characteristic	Category	LSM (SEM) or *β (SE)*	Range	*P*-value
Parity	Primiparous	23.4 (0.87)		<0.01
	Multiparous	17.7 (0.52)		
Milk (kg/d)[2]		*0.40 (0.057)*	11.6-55.0	<0.01
Breeding value persistency		*0.32 (0.12)*	92-113	<0.01

[1] The final multivariable model was based on 23 univariate models. First, cow characteristics were selected at $P < 0.20$ in univariate models, but always with parity in the model. Second, the 9 selected cow characteristics were included in the first multivariable model as independent variables, including interactions with parity, to identify potential predictors for FPCM production end lactation after different VWP. The final model was created by using backward selection at $P < 0.05$.
[2] Average milk production in the final week before successful insemination.

For the prediction of mean BCS in the final 12 wk before dry-off, 13 cow characteristics were selected at $P < 0.20$ from the univariate analyses. These were included in the first multivariable model, next to parity and their interactions with parity. After backward selection, this final multivariable model included 7 cow characteristics (Table 7, figure 5B). With this model, the BCS end lactation was predicted for primiparous cows and multiparous cows separately, as follows:

Primiparous cows:

$$\mathrm{BCS}_{end\ lac} = 6.6 - 0.020 \times \mathrm{MP} + 0.50 \times \mathrm{Prot} - 0.99 \times \mathrm{Lac} + 0.0028 \times \mathrm{BW} + 0.39 \times \mathrm{BCS} - 0.62 \times \mathrm{DMI}_{con},$$

Multiparous cows:

$$\mathrm{BCS}_{end\ lac} = 7.2 - 0.020 \times \mathrm{MP} + 0.50 \times \mathrm{Prot} - 0.99 \times \mathrm{Lac} + 0.0028 \times \mathrm{BW} + 0.39 \times \mathrm{BCS} - 0.62 \times \mathrm{DMI}_{con},$$

where MP is the average milk production in the final week before successful insemination, Prot is the protein content in the final week before successful insemination, Lac is the lactose content in the final week before successful insemination, BW is the average body weight in the final week before successful insemination, BCS is the BCS in the final month before successful insemination, and DMI_{con} is the DMI of concentrate in the first 6 wk of lactation. The residuals in this model were on average 0.3. The adjusted R^2 of the final multivariable model was 0.60, and the adjusted R^2 of the model with VWP, parity, and their interaction (i.e., the model used in table 5) was 0.32.

Table 7. Final multivariable model[1] for prediction of BCS end lactation for cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, VWP200) that completed the first lactation in the experiment and had a dry period (n = 124) (LSM±SEM)

Cow characteristic	Category	LSM (SEM) or *β (SE)*	Range	*P*-value
Parity	Primiparous	2.5 (0.22)		0.04
	Multiparous	3.1 (0.090)		
BCS[2]		*0.39 (0.083)*	1.0 - 3.8	<0.01
Lactose (%)[3]		*-0.99 (0.28)*	4.1 - 5.0	<0.01
BW (kg)[4]		*-2.8 (0.84)* $\times 10^{-3}$	529 - 814	<0.01
DMI concentrate (kg/d)[5]		*-0.62 (0.24)*	4.5 – 6.3	<0.01
Protein (%)[3]		*0.50 (0.19)*	3.0 - 4.6	<0.01
Milk production (kg/d)[6]		*-0.020 (0.0079))*	11.6 - 55.0	0.01

[1] The final multivariable model was based on 23 univariate models. First, cow characteristics were selected at $P < 0.20$ in univariate models, but always with parity in the model. Second, the 13 selected cow characteristics were included in the first multivariable model as independent variables, including interactions with parity, to identify potential predictors for BCS end lactation after different VWP. The final model was created by using backward selection at $P < 0.05$.

[2] Average BCS in the final month before successful insemination.

[3] Content in the final week before successful insemination.

[4] Average BW in the final week before successful insemination.

[5] Average DMI (dry matter intake) of concentrate in the first 6 wk of lactation.

[6] Average milk production in the final week before successful insemination.

For the prediction of FPCM production per day of CInt, 17 cow characteristics were selected at $P < 0.20$ from the univariate analyses. These were included in the first multivariable model, next to parity and their interactions with parity. After backward selection, this final multivariable model included 5 cow characteristics (Table 8, figure 5C). With this model, the FPCM production per day of CInt was predicted for primiparous cows and multiparous cows separately, as follows:

Primiparous cows:

$$\frac{\text{FPCM}}{\text{day CInt}} = -17.4 + 0.25 \times \text{PP} + 0.38 \times \text{MP} + 1.6 \times \text{Fat} + 0.16 \times \text{BV}_{pers},$$

Multiparous cows:

$$\frac{\text{FPCM}}{\text{day CInt}} = -18.9 + 0.25 \times \text{PP} + 0.38 \times \text{MP} + 1.6 \times \text{Fat} + 0.16 \times \text{BV}_{pers},$$

where PP is the peak production between calving and successful insemination, MP is the average milk production in the final week before successful insemination, Fat is the fat content in the final week before successful insemination, and BVpers is the breeding value for persistency. The residuals in this model were on average 1.4 kg FPCM/d. The adjusted R^2 of the final multivariable model was 0.82, and the adjusted R^2 of the model with VWP and parity (i.e., the model used in table 5) was 0.15.

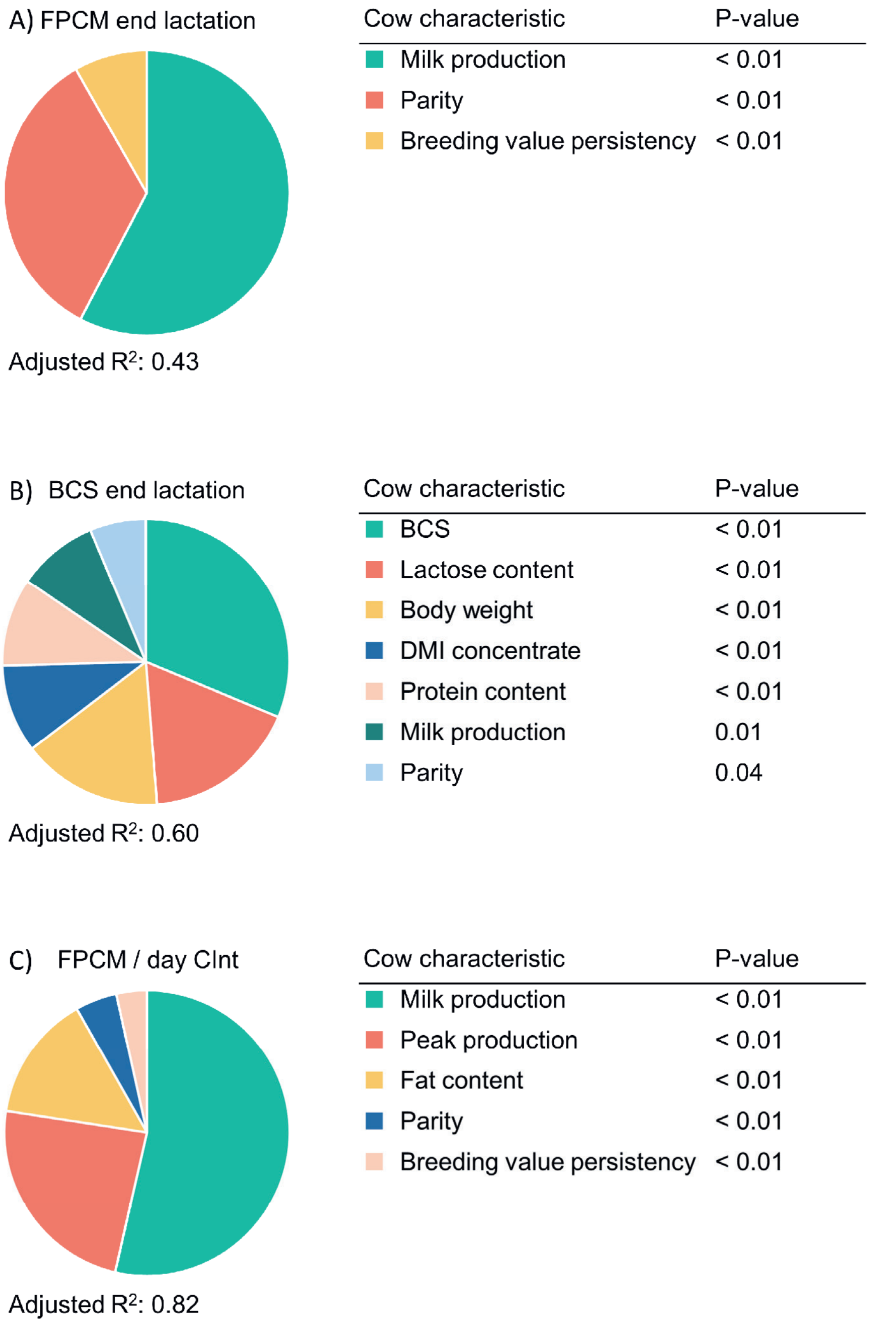
Table 8. Final multivariable model[1] for prediction of fat- and protein-corrected milk (FPCM) per day of calving interval (CInt) for cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, VWP200) that completed the first lactation in the experiment and had a dry period (n = 124) (LSM±SEM)

Cow characteristic	Category	LSM (SEM) or *β (SE)*	Range	*P*-value
Parity	Primiparous	28.2 (0.44)		<0.01
	Multiparous	26.7 (0.24)		
Milk production (kg/d)[2]		*0.38 (0.036)*	11.6 - 55.0	<0.01
Peak production (kg/d)		*0.25 (0.035)*	23.0 - 59.9	<0.01
Fat (%)[3]		*1.7 (0.30)*	3.03 - 4.59	<0.01
Breeding value persistency		*0.16 (0.051)*	92 - 113	<0.01

[1] The final multivariable model was based on 23 univariate models. First, cow characteristics were selected at $P < 0.20$ in univariate models, but always with parity in the model. Second, the 17 selected cow characteristics were included in the first multivariable model as independent variables, including interactions with parity, to identify potential predictors for FPCM production per day of CInt after different VWP. The final model was created by using backward selection at $P < 0.05$.

[2] Average milk production in the final week before successful insemination.

[3] Content in the final week before successful insemination.



A) FPCM end lactation

Cow characteristic	P-value
🟩 Milk production	< 0.01
🟥 Parity	< 0.01
🟨 Breeding value persistency	< 0.01

Adjusted R^2: 0.43

B) BCS end lactation

Cow characteristic	P-value
🟩 BCS	< 0.01
🟥 Lactose content	< 0.01
🟨 Body weight	< 0.01
🟦 DMI concentrate	< 0.01
■ Protein content	< 0.01
■ Milk production	0.01
■ Parity	0.04

Adjusted R^2: 0.60

C) FPCM / day CInt

Cow characteristic	P-value
🟩 Milk production	< 0.01
🟥 Peak production	< 0.01
🟨 Fat content	< 0.01
🟦 Parity	< 0.01
■ Breeding value persistency	< 0.01

Adjusted R^2: 0.82

Figure 5. Cow characteristics that remain in the multivariable model to predict fat- and protein-corrected milk (FPCM) end lactation, BCS end lactation, and FPCM per day of calving interval (CInt) of cows with a voluntary waiting period of 50, 125, or 200 d. Parity always remained in the model. Size of slices are based on type 3 sums of squares.

4 Discussion

This study aimed to investigate how extending the VWP from 50 until 125 or 200 d affected metabolic status, hormones, and body condition development throughout the lactation and the start of the subsequent lactation. When the VWP is extended, insemination takes place later in lactation. At this moment, milk yield can be expected to have decreased (Gaillard et al., 2016). For multiparous cows in the current study, indeed FPCM production and the energy partitioning toward milk at the cost of body reserves both decreased around the end of the VWP when the VWP was increased. Possibly, lower milk production and more energy to BW around the end of an extended VWP are related with an improved fertility. Earlier studies related a lower milk yield to fewer days open after the end of the VWP and greater first service conception rates (Wathes et al., 2007; Niozas et al., 2019b). Indeed, in the current experiment, cows in VWP200 had more normal ovarian cycles (18-24 d in length) around the end of the VWP, and fewer days until pregnancy after the end of the VWP (Ma et al., 2020). Moreover, multiparous cows in VWP200 had greater plasma insulin and IGF-1 concentration around the end of their VWP compared with cows with a shorter VWP. A lower plasma IGF-1 concentration has been related with longer intervals to conception (Wathes et al., 2007). Plasma insulin has been related with improved follicular growth and oocyte development (Fouladi-Nashta et al., 2007; Garnsworthy et al., 2009), although high insulin (> 37.2 µU/mL) was related with a reduced blastocyst yield (Adamiak et al., 2005). The lower milk yield, and less energy partitioned toward milk at the cost of body reserves at start of the insemination period, however, can be hypothesized to contribute to the improved reproductive performance of dairy cows with an extended VWP (Niozas et al., 2019b).

When the end of the VWP was reached, cows were first inseminated when they showed signs of estrus. Estrus may, however, be delayed, and insemination is not always successful (Dobson et al., 2007). Therefore, we also investigated the period around successful insemination. The effect of VWP on metabolites, hormones, and body condition variables was similar for the period around successful insemination compared with the period around the end of the VWP. The only difference with the period around the end of the VWP was that the plasma insulin concentration of multiparous cows did not differ among the 3 VWP groups around successful insemination. One explanation could be that the success of insemination depended on the concentration of insulin, as it did in an earlier study (Garnsworthy et al., 2009). This could indicate that the greater insulin concentration that is related with a delayed insemination could improve insemination success.

A delayed insemination when the VWP is extended also implies that gestation takes place later in lactation. In the current study, in the final stage of the first 305 days the dry period already started for 17 cows in VWP50, while cows in VWP200 could not have been pregnant before 200 days in lactation. Multiparous cows in VWP200 had a greater plasma insulin concentration compared with multiparous cows in VWP50 or VWP125, and a greater plasma IGF-1 concentration compared with multiparous cows in VWP125, during these first 305 days after calving 1. Moreover, primiparous cows in VWP125 had a greater plasma insulin concentration compared with primiparous cows in VWP50. As lactation progresses, insulin increases, whereas start of pregnancy is associated with a decline in insulin (Koprowski and Tucker, 1973). Possibly, this resulted in the lower plasma insulin concentration of cows in shorter VWP during the first 305 days after calving 1. In addition, multiparous cows in VWP200 produced 2.5 kg/d less FPCM and had a lower ratio of NE-FPCM to NE-BW compared with cows in VWP125 during this period. When culled and non-pregnant cows were excluded from the analysis, however, FPCM production and energy partitioning did not differ among the VWP groups in the first 305 days after calving 1. This could indicate that some cows that were diseased or non-fertile or both and later culled lowered the average FPCM production in VWP200. Cows in the VWP200 group that were culled or did not conceive had an average FPCM production of 32.0 kg/d in the first 305 days of lactation. Moreover, when considering only cows that did not conceive or were culled during the first lactation in the experiment, primiparous cows only gained 0.70 kg BW per week, and multiparous cows lost 0.74 kg BW per week in the first 305 days after calving 1. Cows in this group, however, could have been culled before 305 days in lactation.

During the first 305 days after calving 1, the plasma insulin concentration had a relatively great standard error (Figure 1). Visual assessment of individual cows showed that these were not always the same cows, and therefore the great variation in plasma insulin concentration can perhaps be explained by the sampling time. In this study, blood samples were taken 1-3 h after morning milking. For some cows, the time between milking and blood sampling was greater than for other cows. As insulin concentration overall was greater immediately after milking compared with 1 h after milking (Koprowski and Tucker, 1973), this time between milking and sampling could have affected the measurements in the current study. Moreover, more time between milking and blood sampling implies that some cows had more time with access to feed, which could have resulted in an increased plasma insulin concentration for these cows (McAtee and Trenkle, 1971).

As gestation took place later in lactation for cows with an extended VWP, the pregnancy period was at a different time in lactation for the different VWP groups. In this period, multiparous cows in VWP200 had a greater plasma insulin and IGF-1 concentration, greater BCS, greater BW gain, and lowest FPCM production and energy partitioning toward FPCM. Moreover, primiparous cows in VWP125 had a greater plasma insulin concentration compared with primiparous cows in VWP50. Earlier, the IGF-1 and insulin concentration were greater at later compared with earlier lactational stages (Gross et al., 2011). Also, in an earlier study, cows had elevated plasma concentrations of IGF-1, leptin, and glucose, a decreased milk yield, and in increased BW from 301 to 600 DIM compared with from 0 to 300 DIM, indicating partitioning of energy toward body weight later in lactation (Marett et al., 2011). In the current study, gestation was delayed for the extended VWP groups, which could explain these differences in metabolism among the VWP groups.

Moreover, BCS and BW gain before calving could affect the subsequent transition period (Roche and Berry, 2006). Indeed, in the current study, multiparous cows with a VWP of 200 d had a greater BCS, a more negative EB in the first wk after calving, and greater plasma NEFA concentration. This may be related to their greater BW gain and BCS in the end of the previous lactation. Moreover, a more negative EB and greater NEFA concentration in early lactation of these cows could imply that an extended VWP of 200 d in the previous lactation may increase the risk for diseases related with energy balance and metabolic status, such as ketosis and laminitis (Ingvartsen et al., 2003; Friggens et al., 2004).

When cows were grouped for CInt instead of for VWP, similar but more effects were found. After calving 2, multiparous cows in longer CInt also had a more negative EB, a greater BCS, greater plasma NEFA concentration, but next to that a lower DMI, a greater plasma BHB concentration, and a lower plasma insulin concentration compared with cows in shorter CInt. As some cows with a short VWP still had a long CInt, this may have made the effects when comparing CInt groups more profound than when comparing VWP groups (Burgers et al., 2021b). In the current study, ration of cows in the 3 VWP groups was the same, and all cows were fed PMR with concentrates separate in a concentrate feeder. Even though concentrate level was adjusted to milk yield after 100 days in lactation, the basal ration already supplied for approximately 22 kg of milk per day. This might have made it more difficult to keep the cows at a healthy body condition. In practice, farmers could adjust ration of cows that receive a longer VWP to reduce the increase in body condition end of the lactation for cows with an extended lactation. Moreover, especially when a more individual diet composition is not feasible in practice, length of the VWP can be adjusted for individual cows.

In the current study, the effect of VWP on lactation performance was different for multiparous cows and primiparous cows. We did not find an effect of VWP on body condition or FPCM production of primiparous cows. Moreover, in contrast to multiparous cows, for primiparous cows the VWP had no effect on body condition, energy balance, or metabolites in the start of the subsequent lactation. Possibly, this is related to the lack of effect of VWP on milk yield at the end of the previous lactation in primiparous cows (Burgers et al., 2021b) which could have prevented fattening at the end of the lactation. Also, in other studies on extended lactations, primiparous cows maintained their production longer than multiparous cows (Lehmann et al., 2016; Rehn et al., 2000).

To limit the possibility of a reduced milk production at the end of the lactation and related problems in the start of the next lactation, farmers wanted to extend the CInt for specific cows by extending the VWP (Lehmann et al., 2017; Burgers et al., 2021a). Some cows could be better able to maintain their milk production in extended lactations compared with other cows, depending on individual characteristics (Lehmann et al., 2017; Kok et al., 2018). Farmers already used some of these characteristics to select their cows for an extended VWP, such as parity, milk production level, peak production, and body condition (Burgers et al., 2021a). In earlier studies, lactation persistency was estimated for cows with an extended lactation (Manca et al., 2020; Burgers et al., 2021b). Average milk production and maximum milk production in the first 6 wk of lactation and expected 305-d milk production could be used to estimate lactation persistency between day 100 in lactation and dry-off (Burgers et al., 2021b). In the current study, persistency itself was not predicted, as the milk production at the end of the lactation was hypothesized to be more important for performance in extended lactations. Moreover, we used additional information on metabolic status to predict lactation performance. We expressed lactation performance as milk production or BCS at the end of the lactation, or milk production per day of CInt. In commercial dairy practice, definition of a successful extended lactation is subject to each farmer's individual approach and could also include reproductive performance, specific milk components, or simply a reduction in frequency of calving moments or a reduction in number of surplus calves.

In the current study, a greater milk production before successful insemination and a greater breeding value for persistency was related with a greater FPCM production end lactation, which could make cows with a greater milk production before successful insemination and a greater breeding value for persistency more suited for an extended VWP. At the same time, these cows have a greater chance for a high milk production at the end of their lactation when applying a short VWP and with that they might have an increased risk for mastitis during the dry period (Rajala-Schultz et al., 2005). A greater BCS, BW, and protein content before successful insemination was related with an increased BCS end lactation. Earlier, a greater nadir BCS after calving was positively related with protein content in the first 60 days of lactation (Roche et al., 2007). This may explain a relation between protein content in the final week before successful insemination and BCS at the end of the lactation. As an extended VWP in itself increases the risk for an increased BCS end lactation (Niozas et al., 2019a; Burgers et al., 2021b), cows with higher BCS, BW, and protein content before insemination might be less suitable for an extended VWP. A greater milk production and lactose content before insemination was related with a reduced BCS end lactation, and as such cows with a greater milk production and lactose content before insemination could be more suitable for an extended VWP. In the current study, the lactose content before insemination was negatively correlated to the log-transformed SCC before insemination ($r = -0.56$, $P < 0.01$), which may explain the relationship with BCS at the end of the lactation. Possibly, cows with a greater SCC have a reduced milk production, and therefore more risk for fattening at the end of the lactation. Cows with a greater peak production and a greater milk yield before insemination had more chance for a greater FPCM per day of CInt. As such, when these cows are selected for an extended VWP, the negative effect of a longer lactation on milk yield may be limited.

In earlier studies, cows that had lower plasma concentrations of IGF-1, insulin, and glucose between 332 and 620 DIM as a result of a different diet composition could be milked longer (Delany et al., 2010), which could imply that cows with a lower concentration of insulin, IGF-1, and glucose in late lactation or maybe already in early lactation could be more suitable for extended lactations. Moreover, North American Holstein-Friesian cows, compared with New Zealand Holstein-Friesian cows, had lower plasma concentrations of insulin and glucose between 47 and 63 weeks in lactation when both groups received 6 kg concentrate/d, and could be milked longer in a 650-d lactation (Kolver et al., 2007; Kay et al., 2009). These studies indicated that cows with lower concentrations of insulin and IGF-1 partition more energy reserves toward milk production than toward body reserves, resulting in greater lactation persistency. In our study, however, glucose in the first 6 wk and insulin and IGF-1 in the final week before successful insemination did not remain in the multivariate models after backward selection. Possibly, the relationships between metabolites, hormones, energy partitioning, BW, BCS, milk production, milk content, and SCC prevented all these factors together to be selected by the model, so that only the most profound factors remained, in this case mainly milk production and BCS.

This study was performed at one experimental farm, and as such these prediction models are based on the dataset that we used. In practice, when farmers are interested in extending the VWP for part of their herd, they can use these prediction models to only select cows in their herd with a reduced risk for low milk production or fattening at the end of lactation for an extended VWP. Possibly, however, on other farms or with other cows, different cow characteristics could be relevant to predict the FPCM production or the BCS at the end of the lactation, or the FPCM production per day of CInt. Moreover, in our study, only cows that were expected to finish a complete lactation based on being in good general health before the VWP was extended were included. For farmers to be able to select specific cows for an extended VWP, these cow characteristics should be validated for other herds. Despite these limitations of the current data analysis, results are strengthened by the fact that the relations discovered in our study seem biologically sensible, and that similar cow characteristics such as milk production in early lactation and in the previous lactation were relevant in earlier studies (Lehmann et al., 2017). In addition, part of the cow characteristics that we found, such as milk yield before insemination, peak production, and body condition, are already used by farmers for the selection of cows for an extended VWP (Burgers et al., 2021a). This could indicate that the prediction models created in this experiment may point the way for selecting individual cows for an extended VWP in practice.

5 Conclusion

Multiparous cows with an extended VWP had a greater plasma insulin and IGF-1 concentration and a lower FPCM production around the end of the VWP and during pregnancy. Moreover, multiparous cows with an extended VWP gained more BW during pregnancy, which may have resulted in their greater BCS, more negative EB, and greater plasma NEFA concentration at the start of the subsequent lactation. For primiparous cows, the VWP did not affect the body condition or FPCM production during the lactation and the start of the subsequent lactation. Independent of the VWP, a higher milk production and a lower body condition before insemination were associated with a higher milk production and a lower body condition score at the end of the lactation. Variation among cows, especially in parity, milk production and BCS, could call for an individual approach for extending the VWP. Primiparous cows can handle an extended lactation very well. Especially for multiparous cows, an individually customized VWP can prevent fattening end lactation and associated problems in the start of the subsequent lactation, while still having the beneficial effects of a lower frequency of transitions on a herd level.

6 Acknowledgements

The authors thank the staff of the Dairy Campus (Leeuwarden, the Netherlands). This study was financed by DairyNL (ZuivelNL; organization of the Dutch dairy supply chain, The Hague, the Netherlands) and the Dutch Ministry of Agriculture, Nature and Food Quality (LNV, The Hague, the Netherlands) as part of the research program ‘One Health for Food’ (1H4F, The Hague, the Netherlands). The authors state no conflicts of interests.

Appendix

Table A1. The effect (*P*-value) of cow characteristics between calving and successful insemination on the 3 variables for lactation performance: 1. FPCM[1] production in the final 6 wk before dry-off, 2. BCS in the final 12 wk before dry-off, and 3. FPCM production per day of CInt[2], of cows with a voluntary waiting period after calving until first insemination of 50, 125, or 200 days (VWP50, VWP125, VWP200) that completed the first lactation in the experiment and had a dry period (n = 124). All models included parity next to the tested cow characteristic

	Variable for lactation performance		
	1. FPCM end	2. BCS end	3. FPCM / day CInt
First 6 wk			
Energy balance (kJ/$BW^{0.75}$)	0.09	0.58	<0.01
DMI concentrate (kg/d)	0.56	<0.01	0.199
DMI PMR (kg/d)	0.38	0.82	<0.01
NEFA[3] (mmol/L)[4]	0.40	0.6	<0.01
BHB[5] (mmol/L)[4]	0.07	0.18	<0.01
Glucose (mmol/L)[4]	0.45	0.26	<0.01
Between calving and pregnancy			
Peak production (kg milk/d)	0.01	0.49	<0.01
Day of peak production	0.72	0.09	0.87
Slope to peak (kg milk/d)[4,6]	0.26	0.67	<0.01
Slope peak – pregnancy (kg milk/d)[4]	0.46	0.54	0.25
Slope final 3 wk to pregnancy (kg milk/d)	0.69	0.57	0.58
Final week before pregnancy			
Milk production (kg/d)	<0.01	<0.01	<0.01
Fat %	0.31	<0.01	0.12
Protein %	<0.01	<0.01	<0.01
Lactose %	<0.01	<0.01	0.1
Fat : protein ratio	0.52	0.04	0.88
BW (kg)	0.49	<0.01	<0.01
BCS[7]	0.05	<0.01	<0.01
SCC (× 1000)[4]	0.22	0.01	0.28
Insulin (µU/mL)[4]	0.60	0.84	0.30
IGF-1[8] (ng/mL)	<0.01	<0.01	<0.01
Prior data			
Previous 305-d production (kg)[9]	0.31	0.85	<0.01
Breeding value persistency	<0.01	0.03	0.01

[1] FPCM = fat- and protein-corrected milk.
[2] CInt = calving interval.
[3] NEFA = non-esterified fatty acids.
[4] Non-normal data were transformed to approximate a normal distribution.
[5] BHB = β-hydroxybutyrate.
[6] Slope from day 10 in lactation until day of maximum production.
[7] Average in the final month before pregnancy.
[8] IGF-1 = insulin-like growth factor 1.
[9] Previous (multiparous) or expected (primiparous) 305-d milk production.

Chapter 5

Revenues and costs of dairy cows with different voluntary waiting periods based on data of a randomized control trial

E.E.A. Burgers[1,2], A. Kok[1], R.M.A. Goselink[2], H. Hogeveen[3], B. Kemp[1], A.T.M. van Knegsel[1]

[1] Adaptation Physiology Group, Wageningen University & Research, Wageningen, the Netherlands

[2] Wageningen Livestock Research, Wageningen University & Research, Wageningen, the Netherlands

[3] Business Economics Group, Wageningen University & Research, Wageningen, the Netherlands

Journal of Dairy Science 105 (2022): 4171 – 4188

Abstract

Based on modeling studies, a 1-yr calving interval for dairy cows is generally considered optimal from an economic point of view. Recently some dairy farmers are deliberately extending the voluntary waiting period for insemination (VWP) to extend the calving interval. Reasons to extend the VWP are to reduce the frequency of transitions such as dry-off and calving to improve health, to reduce labor associated with these transitions, and to reduce the number of surplus calves. This study aimed to evaluate yearly revenues, yearly costs, and yearly net partial cash flow (NPCF) for individual cows with a VWP of 50, 125, or 200 d based on data from a randomized control trial. The NPCF included revenues and costs for milk yield, calves born, inseminations, concentrate supply, partial mixed ration (PMR) supply, veterinary treatments, discarded milk due to veterinary treatments, culling, and labor (for milking, calving cows, inseminations, and veterinary treatments). Holstein-Friesian dairy cows (n = 153) within one herd were blocked for parity, calving season, and expected (primiparous cows) or previous (multiparous cows) 305-d milk yield. Cows were randomly assigned within the blocks to 1 of 3 VWP (VWP50, VWP125, or VWP200) in wk 6 after calving, and monitored from wk 6 after calving until wk 6 after the next calving or until culling. Revenues and costs were calculated per individual cow and expressed per cow per year. Revenues from milk and costs for PMR and concentrate contributed most to the yearly NPCF. Total yearly revenues were greater in VWP50 compared with VWP200 (€3,169 vs. €2,832), mainly because of €334 greater milk revenues. Total yearly costs were also greater in VWP50 compared with VWP200 (€1,964 vs. €1,729), mainly because of €102 greater concentrate costs. The VWP was not significantly associated with the NPCF per cow per year. A change in milk, feed, or calf price, or a change in labor costs for calving cows or for inseminations had a greater effect on the yearly NPCF of cows in VWP50 compared with cows in VWP200. To investigate variation in NPCF, cows were grouped for yearly NPCF and categorized into 3 economic classes (EC): EC1 (<€1,100/yr), EC2 (€1,100–€1,400/yr), and EC3 (>€1,400/yr). Cows in EC3 had greatest lactation production per day in the experiment (i.e., kg of milk, protein, fat, lactose), and lowest number of veterinary treatments during the experiment.

Key words: extended calving interval, extended lactation, economic result, individual cow variation

1 Introduction

Based on results of recent modeling studies, a 1-yr calving interval (CInt) is generally considered optimal for dairy cows from an economic point of view (Inchaisri et al., 2011; Steeneveld and Hogeveen, 2012; Kok et al., 2019). A longer CInt was associated with a lower milk production per cow per year (Kok et al., 2019). This lower milk production was attributed to the difference in milk yield in early lactation (peak milk yield) and in late lactation (Strandberg and Oltenacu, 1989) and a proportionally longer late-lactation period for cows with an extended lactation. Although a 1-yr CInt seemed optimal from an economic point of view, recently some dairy farmers have been deliberately extending the voluntary waiting period for insemination (VWP; Lehmann et al., 2014; Burgers et al., 2021a). Their motivation is to reduce the frequency of transition periods around dry-off and calving, to reduce the labor associated with these transitions, to reduce the number of surplus calves, and to improve cow health (Lehmann et al., 2014; Burgers et al., 2021a).

Most of the studies on economics of CInt consist of normative simulation modeling, where input is based on retrospective analyses of data from commercial farms (e.g., Strandberg and Oltenacu, 1989; Inchaisri et al., 2011; Kok et al., 2019). In these retrospective analyses, however, a delayed insemination or an extended CInt might be associated with poor fertility and might not necessarily have been the result of a deliberate extension of the VWP in combination with associated management (Mellado et al., 2016). When cows were assigned to a VWP of 60 or 83 d based on their calving dates, total cash flow over a 6-yr period was the same (Gobikrushanth et al., 2014). Moreover, recent experimental studies have assigned cows randomly to a certain VWP, in contrast to the earlier normative modeling studies. When cows were randomly assigned to a VWP of 40 d, 120 d, or 180 d, daily milk yield for the entire lactation was not different (Niozas et al., 2019a), and number of inseminations per pregnant cow was reduced from 1.77 to 1.56 or 1.51 (Niozas et al., 2019b). The BCS at dry-off was increased from 3.25 after a VWP of 40 d to 3.5 after a VWP of 180 d (Niozas et al., 2019a), which could result in an increased risk for diseases after the next calving (Roche et al., 2009). In recent observational studies, where cows were selected and managed for extended CInt, daily milk yield was similar (parity 2 and 3) or greater (parity 1) in a CInt of more than 19 mo compared with shorter CInt (Lehmann et al., 2016). In these farms probably more persistent or more productive cows were selected for increased CInt. Other potential effects on the net cash flow of individual cows after an extended VWP, compared with a shorter VWP, could be lower feed costs for cows in longer lactations (Lehmann et al., 2014); fewer calvings per unit of time per cow, resulting in less risk for calving-related diseases and culling (Fetrow et al., 2006; Pinedo et al., 2014); and fewer calves born. Fewer calves born might reduce revenues from calves, but, due to costs around calving and low revenues from calves, these reductions might not be severe (Mohd Nor et al., 2012).

Therefore, the economic result in a situation with a deliberately extended CInt could differ from the economic result of extended CInt in the retrospective studies. In a randomized control trial, where cows are randomly assigned to a certain VWP, the partial cash flow of individual cows can be calculated for a deliberately extended VWP. Moreover, individual cow characteristics, such as lactation persistency and parity, may influence cow performance in terms of milk yield in an extended CInt (Lehmann et al., 2017; Kok et al., 2019). Calculating cash flows for individual cows could indicate which cow characteristics contribute to individual cow performance after different VWP.

This study aimed to evaluate revenues, costs, and the net partial cash flow for cows that were randomly assigned to a VWP of 50, 125, or 200 d. We investigated complete lactations from wk 6 after calving until wk 6 after the next calving or until culling. The net partial cash flow included revenues and costs for milk yield, calves born, inseminations, concentrate supply, partial mixed ration (PMR) supply, veterinary treatments, discarded milk due to veterinary treatments, and labor (for milking, calving cows, inseminations, and veterinary treatments).

2 Materials and methods

2.1 Animals and housing

The experimental protocol was approved by the Institutional Animal Care and Use Committee of Wageningen University & Research (Wageningen, the Netherlands) and complies with the Dutch law on Animal Experimentation (protocol number 2016.D-0038.005). The experiment was conducted at Dairy Campus Research Farm (Leeuwarden, the Netherlands) between December 2017 and January 2020.

The animals, experimental design, and treatments have been described earlier (Burgers et al., 2021b). In short, 154 cows were selected from a research herd of 500 lactating Holstein-Friesian cows based on the following criteria: no twin pregnancy, no clinical mastitis or SCC >250,000 at the final 2 milk test days before dry-off and expected to finish a complete lactation based on being in good general health. For the current study, cows were followed from wk 6 after calving until wk 6 after the next calving or until culling (experimental days, ED). Cows were milked twice daily around 0600 h and 1800 h in a 40-cow rotary milking parlor (GEA). Partial mixed ration (grass silage, corn silage, soybean meal, wheat meal) supported 22 kg of milk. Concentrate supply started at 1 kg/d on the day of calving and increased stepwise until 21 DIM to 9 kg/d for primiparous cows or 10 kg/d for multiparous cows. After 100 DIM, individual concentrate supply was decreased based on the last 5 d of milk yield. In the milking parlor, 1 kg of additional concentrate was supplied daily. Ration during the dry period consisted of grass silage and corn silage, supplemented with wheat straw and concentrate. In the last 10 d before the expected calving date, cows received 1 kg of concentrate daily. Once per week, cows between 42 and 49 d before the expected calving date were dried off. In the 7 d before dry-off, cows were given the dry cow ration. During the last 3 d before dry-off, cows were milked once daily. When cows had SCC >150,000 cells/mL at the final milk test day, cows were treated with antibiotics at dry-off (Orbenin Dry Cow Extra, Zoetis). All cows were treated with teat sealant at dry-off (Orbeseal, Zoetis).

2.2 Experimental design

The selected 154 animals were blocked for parity, calving date, milk yield in the previous lactation (multiparous cows) or expected milk yield (primiparous cows), and breeding value for persistency (CRV, Arnhem, the Netherlands) wk 6 after calving. Each block consisted of 3 cows. First, 50 blocks of 3 cows were formed. After removal of 2 cows before the end of the VWP due to culling as a result of health issues, 2 more blocks of 3 cows were added. Per block, the cows were randomly divided over 3 treatment groups: a VWP of 50 d (VWP50), 125 d (VWP125), or 200 d (VWP200). Cows in the 3 treatment groups were inseminated after their VWP when estrus was detected. Estrus detection was carried out by using neck-mounted 3-dimensional accelerometers (Nedap Smarttag Neck) in combination with visual observations by the animal caretaker. Cows were inseminated until 300 DIM. Cows that did not conceive within 300 DIM stayed in the experiment until 530 DIM as long as they produced at least 10 L of milk per day.

2.3 Measurements

Milk yield (MY) was recorded at every milking, from wk 6 after calving until dry-off, and the first 6 wk of the next lactation. Milk samples for the analysis of fat, protein, and lactose were collected for each individual cow from the container 4 times per week (Tuesday afternoon, Wednesday morning, Wednesday afternoon, Thursday morning) in 10-mL tubes containing Bronopol as a preservative and analyzed for the percentage of fat, protein, and lactose as a pooled sample (ISO, 2013; Qlip, Zutphen, the Netherlands). Body weight was recorded twice daily after milking, using a scale that the cows walked over when returning from the milking rotary to the pen (GEA). Body condition score was visually evaluated every 4 wk by the same person, using a 1-to-5 scale (Ferguson et al., 1994). Concentrate supply was recorded for all individual cows in individual feeding boxes. Inseminations were recorded for all individual cows, including cows that did not conceive (in 300 DIM). Veterinary treatments were recorded for all individual cows and included all preventive and curative treatments that cows received (Appendix Table A1).

2.4 Economic calculations

For this study, we defined the net partial cash flow (NPCF) as the result of cash inflows and outflows associated with the calving interval and insemination decisions. More specifically, the NPCF of an individual cow k included revenues from milk ($\mathrm{R}^{\mathrm{MILK}}$) and calves ($\mathrm{R}^{\mathrm{CALVES}}$), and costs for inseminations ($\mathrm{C}^{\mathrm{INS}}$), concentrate ($\mathrm{C}^{\mathrm{CON}}$), PMR ($\mathrm{C}^{\mathrm{PMR}}$), veterinary treatments ($\mathrm{C}^{\mathrm{VET}}$), and culling ($\mathrm{C}^{\mathrm{CULLING}}$) during the experiment:

$$\mathrm{NPCF}_k = \left(\mathrm{R}_k^{\mathrm{MILK}} + \mathrm{R}_k^{\mathrm{CALVES}}\right) - \left(\mathrm{C}_k^{\mathrm{INS}} + \mathrm{C}_k^{\mathrm{CON}} + \mathrm{C}_k^{\mathrm{PMR}} + \mathrm{C}_k^{\mathrm{VET}} + \mathrm{C}_k^{\mathrm{CULLING}}\right).$$

Next, the NPCF_k was expressed per year, by dividing over the experimental days (ED):

$$\mathrm{NPCF}_k^{\mathrm{YEAR}} = \frac{\mathrm{NPCF}_k}{\mathrm{ED}_k} \times 365.$$

Finally, the NPCF_k was aggregated per VWP × parity class and expressed per year, as follows:

$$\mathrm{NPCF}_{ij}^{\mathrm{VWP,ParityClass}} = \frac{\Sigma_{k=1}^{n}(\mathrm{NPCF}_k)}{\Sigma_{k=1}^{n}(\mathrm{ED}_k)} \times 365,$$

where i represents the VWP (i = 50, 125, or 200 d), and j represents the parity class (j = primiparous or multiparous). This weighted mean was calculated to account for the number of ED of each individual cow. Consequently, cows that spent less time in the experiment had a smaller contribution to the weighted NPCF per cow per year.

Milk, protein, fat, and lactose

Milk yield was averaged per week and summed for the complete experiment per cow. From the weekly milk samples, content was determined, and thus protein yield (PY), fat yield (FY), and lactose yield (LY) in kg/d were calculated per week and summed for the complete experiment per cow. With this, total milk revenues ($\mathrm{R^{MILK}}$) were calculated per individual cow k, as follows:

$$\mathrm{R}_k^{\mathrm{MILK}} = (\mathrm{P}^{\mathrm{MILK\,VOL}} \times \mathrm{MY}_k) + (\mathrm{P}^{\mathrm{PROT}} \times \mathrm{PY}_k) + (\mathrm{P}^{\mathrm{FAT}} \times \mathrm{FY}_k) + (\mathrm{P}^{\mathrm{LACT}} \times \mathrm{LY}_k) - \mathrm{P}_k^{\mathrm{LABOR}},$$

where $\mathrm{P^{MILK\ VOL}}$ is the price for the milk volume, $\mathrm{P^{PROT}}$ is the price per kilogram of protein, $\mathrm{P^{FAT}}$ is the price per kilogram of fat, $\mathrm{P^{LACT}}$ is the price per kilogram of lactose, and $\mathrm{P^{LABOR}}$ is the price for milking labor of cow k.

Calves

For cows that had a second calf within the experiment, revenues from calves ($\mathrm{R^{CALVES}}$) were calculated per individual cow k, as follows:

$$\mathrm{R}_k^{\mathrm{CALVES}} = \mathrm{P}^{\mathrm{CALF}} - (\mathrm{P}^{\mathrm{MILK\,REPLACER}} + \mathrm{P}^{\mathrm{LABOR}}),$$

where $\mathrm{P^{CALF}}$ is the price for a calf, $\mathrm{P^{MILK\ REPLACER}}$ is the price for milk replacer consumption of the calf, and $\mathrm{P^{LABOR}}$ is the labor price for a calving cow.

Inseminations

Costs for insemination ($\mathrm{C^{INS}}$) were calculated per individual cow k, as follows:

$$\mathrm{C}_k^{\mathrm{INS}} = (\mathrm{P}^{\mathrm{INS}} + \mathrm{P}^{\mathrm{LABOR}}) \times \mathrm{INS}_k,$$

where $\mathrm{P^{INS}}$ is the semen price, $\mathrm{P^{LABOR}}$ is the labor price per insemination, and INS is the number of inseminations per cow k.

Feed

Costs for feed were calculated both in the dry period and in the lactating periods. Costs for concentrate supply (C^{CON}) were calculated per individual cow *k*, as follows:

$$\mathrm{C}_k^{\mathrm{CON}} = \mathrm{P}^{\mathrm{CON}} \times \mathrm{CONSUP}_k,$$

where P^{CON} is the price per kilogram of concentrate, and CONSUP is the total amount of concentrate (kg) supplied to cow *k* in the complete experiment. The concentrate supply consists of both concentrate in feeders with individual concentrate supply and concentrate in the milking parlor.

Costs for PMR supply (C^{PMR}) were calculated per individual cow *k*, as follows:

$$\mathrm{C}_k^{\mathrm{PMR}} = \mathrm{P}^{\mathrm{PMR}} \times \mathrm{PMRSUP}_k,$$

where P^{PMR} is the price per kilogram of PMR, and PMRSUP is the total amount of PMR (kg) supplied to cow *k* in the complete experiment. To estimate PMR supply, energy requirements for maintenance, milk production, growth, and gestation were calculated per individual cow, using the Dutch VEM system (Feed Unit Milk; 1,000 VEM = 6.9 MJ of NE) with requirements for maintenance and FPCM. For this calculation, milk production was converted to FPCM using the following formula (CVB, 2016):

$$\mathrm{FPCM}_k(\mathrm{kg}) = \mathrm{MY}_k(\mathrm{kg}) \times (0.337 + 0.116 \times \mathrm{fat}_k(\%) + 0.06 \times \mathrm{protein}_k(\%)),$$

after which the energy requirements were calculated per cow *k* (Dutch net energy system for lactation; CVB, 2016):

$$\mathrm{VEM}_k = \left(42.2 \times \mathrm{BW}_k^{0.75} + (442 \times \mathrm{FPCM}_k)\right) \times (1 + (\mathrm{FPCM}_k - 15) \times 0.00165) + \mathrm{VEM}_k^{\mathrm{GROWTH}} + \mathrm{VEM}_k^{\mathrm{GEST}},$$

where $BW^{0.75}$ is the metabolic BW of cow *k*, VEM^{GROWTH} are the energy requirements for growth, and VEM^{GEST} are the energy requirements for gestation. After calculating the total energy requirements of each cow, energy from concentrate supply was subtracted to estimate energy from PMR supply. Using the mean VEM of the PMR in the experiment (437 VEM per kg of product), PMR supply in kilograms was estimated.

Veterinary treatments

Costs for veterinary treatments ($\mathrm{C}^{\mathrm{VET}}$) were calculated per individual cow *k* for the complete experiment, as follows:

$$\mathrm{C}_k^{\mathrm{VET}} = \Sigma_{l=1}^{n}\left(\left(\mathrm{P}_l^{\mathrm{MEDICINE}} + \mathrm{P}_l^{\mathrm{LABOR}}\right) + \left(\mathrm{WP}_l \times \mathrm{MY}_k^{\mathrm{WP}} \times \mathrm{P}^{\mathrm{DISC\ MILK}}\right)\right),$$

where *n* is the number of veterinary treatments for cow *k*, $\mathrm{P}^{\mathrm{MEDICINE}}$ is the price for medicine *l* (Appendix Table A1), $\mathrm{P}^{\mathrm{LABOR}}$ is the labor price for medicine *l*, WP_l is the waiting period in days for medicine *l*, $\mathrm{MY}^{\mathrm{WP}}$ is the milk yield (kg/d) of cow *k* during the waiting period, and $\mathrm{P}^{\mathrm{DISC\ MILK}}$ is the price for the discarded milk per kilogram.

Culling

Costs for culling ($\mathrm{C}^{\mathrm{CULLING}}$) were calculated per individual cow that was culled due to health issues, or that did not become pregnant within 300 DIM in the first lactation in the experiment, as follows:

$$\mathrm{C}_k^{\mathrm{CULLING}} = \frac{\mathrm{C}^{\mathrm{REARING}} - \left(\mathrm{P}^{\mathrm{SLAUGHTER}} \times (0.6 \times \mathrm{BW}_k)\right)}{\text{AIMED PRODUCTIVE DAYS}} \times \left(\text{AIMED PRODUCITVE DAYS} - \left(\text{CULLING AGE}_k - \text{REARING DAYS}\right)\right),$$

where $\mathrm{C}^{\mathrm{REARING}}$ are the rearing costs, $\mathrm{P}^{\mathrm{SLAUGHTER}}$ is the slaughter price per kilogram, BW_k is the body weight of a cow *k* at the time of culling, 0.6 is the dressing percentage of a cow (60%; Rutten et al., 2014), and culling age is the age at culling of cow *k* in days.

For analysis, revenues (R) and costs (C) for all different variables were expressed per cow *k* per year, as follows:

$$\mathrm{R}_k^{\mathrm{YEAR}} \text{ or } \mathrm{C}_k^{\mathrm{YEAR}} = \frac{\mathrm{R}_k \text{ or } \mathrm{C}_k}{\mathrm{ED}_k} \times 365.$$

2.5 Input

For the calculations, some input was assumed based on the Dutch dairy farming system between 2015 and 2020 (Table 1). Moreover, information from the network of dairy farmers (Burgers et al., 2021a) was used for input on labor costs for milking and for calving cows. Prices for milk were calculated as the average price for protein, fat, and lactose from FrieslandCampina (2020) between 2015 and 2020. Costs for milking were calculated assuming 100 cows per hour were milked in the 40-cow rotary milking parlor (GEA), and 1 h of own labor was worth €26 (mean from network dairy farmers). The price for calves was calculated assuming that the chance for a bull or a heifer is 50%, rearing costs for calves were calculated assuming a total milk replacer consumption of 6 L/d (9 kg of milk replacer) in the first 14 d except for the first 2 d (KWIN-V, 2020), and labor costs for a calving cow were assumed to be €36 (mean from network dairy farmers). The costs for 1 insemination were based on labor costs for insemination (KWIN-V, 2020) and the semen price (KWIN-V, 2020). To calculate feed costs, energy requirements for growth (VEM^{GROWTH}) were assumed to be 660 VEM/d for cows that were between 2 and 3 yr of age, and 330 VEM/d for cows that were between 3 and 4 yr of age (Kok et al., 2019). Energy requirements for gestation (VEM^{GEST}) started from 5 mo of pregnancy and were assumed to be in total 167,750 VEM (CVB, 2016). This was added for cows that had a second calf within the experiment. Costs for veterinary treatments were based on information from the veterinarian of University Livestock Practice (Harmelen, the Netherlands). Losses for discarded milk were calculated for the waiting period per treatment with the MY of the cow at that moment. For each cow that was culled due to health issues, revenues from slaughter were calculated with the slaughter value per kilogram of slaughter weight. For this calculation, individual body weight at the time of culling was used. Culling costs were calculated assuming a depreciation method with an aimed lifespan of 8 yr (2,920 d) and lactation starting at 2 yr of age (730 d), resulting in 6 productive years (2,190 d). Moreover, costs for rearing were assumed to be €1,567 per cow (Mohd Nor et al., 2012). Non-pregnant cows left the experiment at approximately 530 DIM and were assumed to be culled at that moment for the calculation of culling costs.

Table 1. Monetary value (€) used to calculate the economic result of cows that had a voluntary waiting period of 50, 125, or 200 days

Variable		Reference
Milk		
Milk (€/100 kg)	-0.67	FrieslandCampina (2015-2020)
Protein (€/100 kg)	550.84	FrieslandCampina (2015-2020)
Fat (€/100 kg)	275.42	FrieslandCampina (2015-2020)
Lactose (€/100 kg)	55.08	FrieslandCampina (2015-2020)
Milking labor costs (€/cow per day)	0.52	GEA, Dusseldorf, Germany Mean of dairy farmers network
Calves		
Milk replacer first 14 days[1] (€/calf)	18	KWIN, 2020
Labor around a calving cow (€/calf)	36	Mean of dairy farmers network
Heifer sold (€/calf)	25	KWIN, 2020
Bull sold (€/calf)	105	KWIN, 2020
Inseminations		
Labor (€/insemination)	13.75	KWIN, 2020
Semen (€/straw)	18	KWIN, 2020
Feed		
Concentrate (€/100 kg)[2]	26	Dairy Campus (2018-2019)
Partially mixed ration (PMR) (€/100 kg)[3]	9.39	Dairy Campus (2018-2019)
Veterinary treatments		
Veterinary treatments (€/treatment)	Table A1	Veterinarian Dairy Campus
Labor preventive treatment (€)	5	Veterinarian Dairy Campus
Labor simple treatment (€)	15	Veterinarian Dairy Campus
Labor multiple treatments (€)	30	Veterinarian Dairy Campus
Discarded milk (€/100 kg)	33.7	Mean price FrieslandCampina (2015-2020)
Culling		
Rearing costs	1,567	Mohd Nor et al., 2012
Slaughter value (€/kg slaughter weight)	2.01	Rutten et al., 2014

[1] 6 L / day except for first 2 days; in total 72 L (9 kg milk replacer).

[2] Mean price for concentrate Dairy Campus 2018 – 2019.

[3] Mean price for PMR (grass silage, corn silage, soy, wheat) Dairy Campus 2018 – 2019.

2.6 Statistical analysis

One cow was culled on d 43 after calving; therefore 153 cows were included in the analyses (41 primiparous and 112 multiparous cows). Parity class (primiparous or multiparous cows) refers to the parity of the cow during the first lactation within the experiment. Statistical analyses were performed using SAS version 9.4 (SAS Institute Inc.). Values are presented as least squares means (LSM) ± standard error of the mean. All *P*-values of pairwise comparisons of LSM were corrected with a Bonferroni adjustment. Two data sets were used for the analyses. Data set 1 included all cows and was used to investigate mean output per cow for their time in the total experiment. Data set 2 included only cows of which the yearly culling costs did not exceed twice the standard deviation of yearly culling costs. As a result, 1 cow from VWP50 and 2 cows from VWP200 were excluded from this data set, as they were culled relatively early in lactation (at respectively 56, 69, and 69 DIM), leading to very high costs of culling expressed per year. The high yearly culling costs were caused by the short time of these cows in the experiment.

Data set 1 (n = 153)

Data set 1 was used to investigate mean output per cow for the total ED. A general linear mixed model (PROC MIXED) was used to test the effects of VWP, parity class, and the interaction between VWP and parity class on the dependent variables: days to pregnancy, days to pregnancy after end of VWP, calving to first service interval (CFSI, d), CInt (d), ED (d), dry period length (d), total MY (kg), total PY (kg), total FY (kg), total LY (kg), total concentrate supply (kg), or total PMR supply (kg). Non-significant interactions ($P > 0.05$) were removed from the models. A logistic regression model with a binary distribution (PROC LOGISTIC) was used to model the probability that a calf was born, that a cow had a veterinary treatment, that milk of a cow was discarded due to veterinary treatment, that a cow was culled due to health issues, or that a cow did not become pregnant within 300 DIM, with fixed effects of VWP and parity class. For cows that had at least 1 veterinary treatment, a generalized linear mixed model with a negative binomial distribution (PROC GLIMMIX) was used to test the effects of VWP, parity class, and the interaction between VWP and parity class on the number of veterinary treatments. For cows of which milk was discarded, a general linear mixed model (PROC MIXED) was used to test the effects of VWP, parity class, and the interaction between VWP and parity class on the discarded milk, where a square root transformation of the discarded milk was used to approximate a normal distribution. A generalized linear mixed model with a

Poisson distribution (PROC GLIMMIX) was used to test the effects of VWP, parity class, and the interaction between VWP and parity class on the total number of inseminations. A general linear mixed model (PROC MIXED) was used to test the effects of VWP, parity class, and the interaction between VWP and parity class on total NPCF per cow. Moreover, data set 1 was used to calculate the weighted NPCF per cow per year.

Data set 2 (n = 150)

Data set 2 was used to investigate revenues, costs, and NPCF per cow per year. In this data set, cows with yearly culling costs that exceeded twice the standard deviation of yearly culling costs were excluded. A general linear mixed model (PROC MIXED) was used to test the effects of VWP, parity class, and the interaction between VWP and parity class on the revenues and costs for milk, calves, PMR supply, concentrate supply, inseminations, veterinary treatments, culling due to health issues, not becoming pregnant, and NPCF per cow per year. Non-significant interactions were removed from all models. A log-transformation of the costs for veterinary treatments was used to approximate a normal distribution. For the analysis of calf revenues, only cows that had a second calf in the experiment were included (n = 127). For the analysis of costs for culling due to health issues, only cows that were culled due to health issues were included (n = 15). Similarly, for the analysis of costs for not becoming pregnant, only cows that did not become pregnant were included (n = 14). Statistical analyses were not performed on culling costs or costs for not becoming pregnant for all cows, as these data were zero inflated. However, to provide complete information, we did calculate the mean culling costs and mean costs for not becoming pregnant for all cows, including cows that had no costs for culling or not becoming pregnant. Data set 2 was also used to calculate the weighted NPCF per cow per year.

Moreover, data set 2 was used to investigate individual variation in NPCF per cow per year. Cows were grouped for yearly NPCF and categorized into 3 economic classes (EC), based on similar numbers of cows per class: EC1 (<€1,100/yr; n = 50), EC2 (€1,100–€1,400/yr; n = 51), and EC3 (>€1,400/yr; n = 49). First, a chi-squared test was used to assess whether VWP determined in which EC a cow was categorized (PROC FREQ). Second, a general linear mixed model (PROC MIXED) was used to test the effect of EC, VWP, parity class, and all 2-way interactions on several dependent variables within the experimental period from wk 6 after calving until wk 6 after the next calving: CFSI, CInt, lactation persistency between d 100 and start of dry-off, MY, PY, FY, and LY per ED, mean content of protein, fat, and lactose, veterinary treatments (log-transformed to approximate a normal distribution), mean BW, mean BCS, and NPCF. Non-significant interactions ($P > 0.05$) were removed from all models.

Finally, cow characteristics from the first 6 wk after the first calving within the experiment (as described in Burgers et al., 2021b) were added to data set 2. These cow characteristics were used to evaluate which characteristics in early lactation could be used to predict yearly NPCF of cows after different VWP. The following cow characteristics in early lactation (first 6 wk) were tested: maximum yield, day of maximum yield, slope to maximum yield, mean MY, mean FPCM yield, fat, protein, and lactose contents, fat-to-protein ratio, BCS, and BW. Next to these early-lactation characteristics, expected (primiparous cows) or previous (multiparous cows) 305-d MY and breeding value for persistency were tested. First, the effect of each cow characteristic on yearly NPCF was tested with a univariate analysis, using a general linear mixed model in SAS (PROC MIXED). Second, when P-value was <0.2, the characteristic was included in the multivariate model. The multivariate model always included VWP and parity class as fixed effects. The cow characteristics in early lactation and their interaction with VWP and parity class stayed in the model if $P < 0.05$, using backward selection.

2.7 Sensitivity analysis

A sensitivity analysis was performed to assess the effect of changes in revenues and costs on the NPCF per cow per year of primiparous and multiparous cows (data set 2, n = 150). First, revenues for milk were based on either the lowest monthly price for protein, fat, and lactose (€408, 204, and 41 per 100 kg) or the highest monthly price for protein, fat, and lactose (€682, 341, and 68 per 100 kg) between 2015 and 2020 (FrieslandCampina, 2020). Second, costs for concentrate and basal ration were based on either the lowest price (concentrate: 20.6; basal ration: €8.25/100 kg) or the highest price for these (concentrate: 26.4; basal ration: €9.5/100 kg) between 2015 and 2020 (Agrimatie; KWIN-V, 2020). Third, calf prices were either €0 or €130 per calf (double value). Fourth, labor costs for calving cows were either €1.25 or €100 per calf (minimum and maximum from network dairy farmers). Fifth, labor costs for inseminating cows were either €0 or €27.5 per insemination. Sixth, labor costs for veterinary treatments were either €0 or the maximum labor costs (€30) per veterinary treatment.

A general linear mixed model (PROC MIXED) was used to test the effects of VWP, parity class, and the interaction between VWP and parity class on the change in yearly NPCF as a result of minimum and maximum milk prices, minimum and maximum feed prices, minimum and maximum calf prices, minimum and maximum labor costs for calving cows (log-transformed to approximate a normal distribution), minimum and maximum labor costs for inseminations, and minimum and maximum labor costs for veterinary treatments (log-transformed to approximate a normal distribution). Nonsignificant interactions ($P > 0.05$) were removed from the models.

3 Results

Of the 153 cows that entered the experiment, 127 cows had a second calf, and 121 cows finished the complete experiment from wk 6 after calving until wk 6 after the next calving. In total, 14 cows did not become pregnant within 300 DIM during the first lactation (2 from VWP50, 3 from VWP125, 9 from VWP200), and 18 cows were culled during the study due to health issues (4 from VWP50, 7 from VWP125, 7 from VWP200). One cow from VWP50 was successfully inseminated at 48 DIM and remained in the study. Increasing the VWP resulted in a greater CFSI [67 (48–113) vs. 140 (125–174) vs. 210 (200–225) d for VWP50, VWP125, and VWP200, $P < 0.01$] and greater calving interval (382 vs. 450 vs. 498 d for VWP50, VWP125, and VWP200, $P < 0.01$). Consequently, cows in VWP200 and VWP125 had more ED compared with cows in VWP50 (470 and 437 vs. 365 d, $P < 0.01$). Increasing the VWP resulted in more days to pregnancy after calving (104 vs. 172 vs. 221 for VWP50, VWP125, and VWP200, $P < 0.01$). Cows in VWP200 had fewer days to pregnancy after end of the VWP (21 d) compared with cows in VWP50 (54 d, $P < 0.01$) or compared with cows in VWP125 (47 d, $P = 0.04$). Dry period length did not differ among the 3 VWP.

3.1 Total output per cow in the complete experiment

The VWP affected total kilograms of milk, fat, protein, and lactose, and total PMR supply in the complete experiment (Table 2). Cows in VWP200 had on average 54 kg greater PY (P = 0.03) and 88 kg greater FY ($P < 0.01$) and tended to have 1,305 kg greater MY (P = 0.07) in the complete experiment compared with cows in VWP50. Cows in VWP125 had on average 79 kg greater FY ($P < 0.01$) and tended to have 1,367 kg greater MY (P = 0.06), 49 kg greater PY (P = 0.06), and 59 kg greater LY (P = 0.08) in the complete experiment compared with cows in VWP50. In the complete experiment, on average 2,968 kg more PMR was supplied to cows in VWP200 ($P < 0.01$), and on average 2,194 kg more PMR was supplied to cows in VWP125 ($P < 0.04$), compared with cows in VWP50 (Table 2). The VWP tended to affect the probability that a cow did not become pregnant, but no statistically significant differences existed among the 3 VWP groups. In the complete experiment, the VWP did not affect total number of inseminations, concentrate supply, probability and number of veterinary treatments, probability and amount of discarded milk, or probability of culling due to health issues. The net partial cash flow in the complete experiment was first assessed for primiparous cows and multiparous cows separately (Figure 1). The VWP or parity class did not affect the NPCF in the complete experiment.

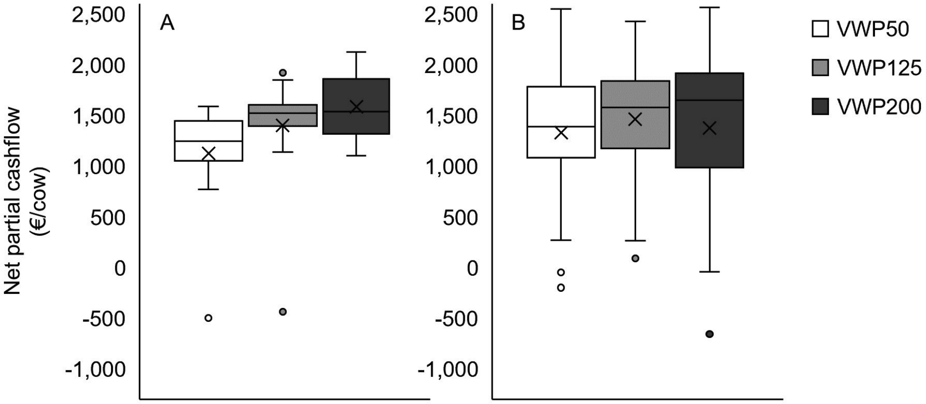


Figure 1. Net partial cash flow (€/cow) in the complete experimental period for primiparous (A) and multiparous (B) cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, VWP200) (N = 153). The × in the box indicates the mean, the solid black line in the box indicates the median, top and bottom of the box are the first and the third quartiles, and circles indicate the outliers.

Table 2. Cow performance for the complete experiment from wk 6 after calving until wk 6 after the next calving or until culling for primiparous and multiparous cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, VWP200) (LSM±SEM) (N = 153)

	Voluntary waiting period						Parity class				*P*-value[1]	
	VWP50		VWP125		VWP200		Primiparous cows		Multiparous cows		VWP	Par
	LSM	SEM (CI)	LSM	SEM (CI)	LSM	SEM (CI)	LSM	SEM (CI)	LSM	SEM (CI)		
n cows	53		49		51		41		112			
CFSI[2] (d)	67^{c}	2	140^{b}	2	210^{a}	2	139	2	139	1	<0.01	0.97
Calving interval[3] (d)	382^{c}	7	450^{b}	7	498^{a}	8	438	8	449	5	<0.01	0.25
Experimental days[4] (d)	365^{c}	13	437^{b}	14	470^{a}	14	426	15	423	9	<0.01	0.86
Dry period (d)	40	1	41	2	42	2	40	2	42	1	0.73	0.15
Milk (kg)	9,820†‡	422	11,187†	431	11,125‡	434	10,107	458	11,315	277	0.03	0.03
Protein (kg)	359^{b†}	15	408^{ab†}	16	413^{a}	16	375	17	412	10	0.02	0.06
Fat (kg)	421^{b}	18	500^{a}	19	509^{a}	19	462	20	492	12	<0.01	0.19
Lactose (kg)	446†	19	505†	19	500	20	462	21	505	13	0.047	0.08
Calves (number)	0.9	0.0	0.9	0.0	0.8	0.1	0.9	0.1	0.8	0.0	0.15	0.38
Inseminations (number)	2.1	0.2	2.4	0.2	1.8	0.2	2.0	0.2	2.2	0.1	0.17	0.46
Concentrate (kg)	1,837	78	2,015	80	1,920	81	1,783	85	2,065	52	0.26	<0.01
Partially mixed ration (kg)	11,990^{b}	484	14,184^{a}	495	14,958^{a}	498	13,440	525	13,981	318	<0.01	0.38
Veterinary treatments (%)[5]	100	0	96	3	93	4	100	11	100	6	0.90	0.48
Veterinary treatments (number)[6]	6.1	0.8	7.3	1.0	6.4	0.9	4.3	0.6	10.2	0.8	0.57	<0.01
Discarded milk (%)[7]	47	8	38	7	38	7	27	7	57	5	0.58	<0.01
Discarded milk (kg)[8]	246	(146-371)	286	(167-436)	257	(150-392)	234	(108-408)	293	(227-367)	0.87	0.49
Culling due to health issues (%)[9]	5.3	3.0	10.7	4.7	9.6	4.5	4.7	3.2	14.0	3.3	0.49	0.12
Not becoming pregnant (%)[10]	3.5	2.5	5.8	3.4	16.7	5.8	6.3	3.7	8.0	2.7	0.05	0.71
Net partial cash flow (€)	1,268	91	1,437	93	1,416	94	1,360	99	1,388	60	0.34	0.81

[a-c] Different superscript indicates a difference among LSM from VWP or parity class ($P < 0.05$).

[1] VWP = voluntary waiting period. Par = parity (primiparous or multiparous) in the first lactation in the experiment. Interaction VWP × parity class was never significant and therefore not included in the models.

[2] CFSI = calving to first service interval: interval from the first calving within the experiment until first insemination.

[3] Calving interval is presented for cows that had a second calf within the experiment (n = 127).

[4] Experimental days: number of days from wk 6 after calving until wk 6 after the next calving, so including the dry period, or until culling (N = 153).

[5] Probability of at least one veterinary treatment per cow.

[6] If veterinary treatment occurred (n = 148): number of veterinary treatments.

[7] Probability of at least once discarded milk as a consequence of a veterinary treatment.

[8] If milk was discarded (n = 75): amount of discarded milk. *P*-values based on square root transformation, data are back-transformed, and confidence interval (CI) is shown.

[9] Probability that a cow was culled due to health issues.

[10] Probability that a cow did not become pregnant within 300 DIM.

[†‡] Similar symbol indicates a trend in difference among LSM from VWP or parity class ($P < 0.10$).

3.2 Yearly revenues and costs per cow

Mean yearly revenues and costs were first assessed for primiparous cows and multiparous cows separately (Figure 2). Yearly revenues from milk and yearly costs for PMR were the greatest contributors to the yearly NPCF in the 3 VWP. Yearly revenues from calves and yearly costs for inseminations were relatively small and therefore not visible in the figure.

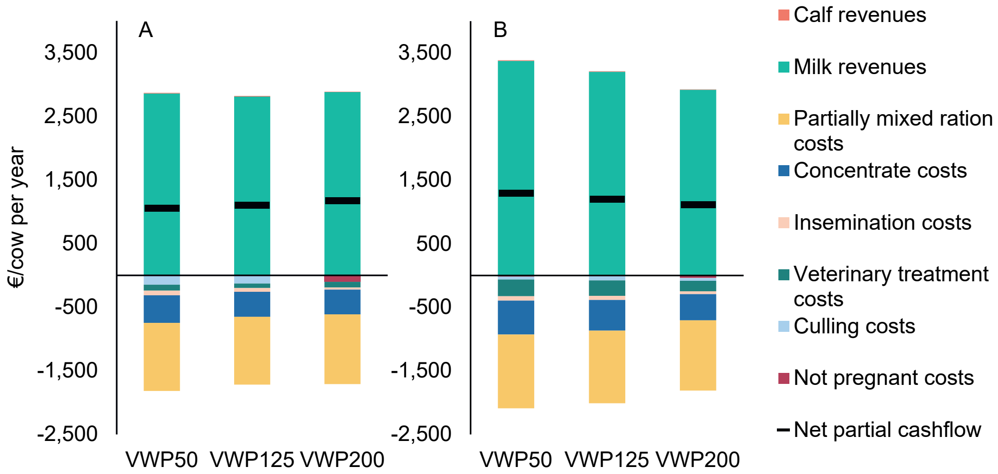


Figure 2. Revenues, costs, and net partial cash flow (NPCF) per cow year for primiparous (A) and multiparous (B) cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, VWP200) (n = 150).

In the analysis of yearly revenues and costs per cow, the interaction between VWP and parity class was never significant and was therefore taken out of the models. The VWP affected both total yearly revenues and total yearly costs (Table 3). Cows in VWP50 had on average €337 greater yearly revenues compared with cows in VWP200 ($P < 0.01$), mainly because of €334 greater yearly milk revenues ($P < 0.01$). Moreover, yearly calf revenues were greatest for cows in VWP50, intermediate for cows in VWP125, and lowest for cows in VWP200 ($P < 0.01$). Cows in VWP50 had on average €235 greater total yearly costs compared with cows in VWP200 ($P = 0.02$), mainly because of €102 greater yearly concentrate costs ($P < 0.01$). Moreover, cows in VWP200 had on average €28 lower yearly inseminations costs compared with VWP50 ($P < 0.01$), and on average €22 lower yearly insemination costs compared with VWP125 ($P = 0.03$). The costs of not becoming pregnant did not differ among VWP, and the VWP did not affect yearly costs per cow for PMR, veterinary treatments, or culling due to health issues. Moreover, the VWP was not significantly associated with the NPCF per cow per year, but numerically the NPCF per cow per year was €55/cow per year greater for cows in VWP50 compared with cows in VWP125 and €102/cow per year greater for cows in VWP50 compared with cows in VWP200. The weighted NPCF per cow per year was numerically €71/cow per year greater for cows in VWP50 compared with cows in VWP125 and €168/cow per year greater for cows in VWP50 compared with cows in VWP200.

Table 3. Revenues and costs (€/cow per year) for milk, calves, partially mixed ration supply, concentrate supply, veterinary treatments[1], culling, and inseminations, and the net partial cash flow (NPCF; €/cow per year) per cow for primiparous and multiparous cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, VWP200) (LSM±SEM) (n = 150)

	Voluntary waiting period						Parity class				*P*-value[2]	
	VWP50		VWP125		VWP200		Primiparous cows		Multiparous cows		VWP	Par
	LSM	SEM (CI)	LSM	SEM (CI)	LSM	SEM (CI)	LSM	SEM (CI)	LSM	SEM (CI)		
n cows	52		49		49		41		109			
Revenues[3]	$3,169^{a}$	68	$3,027^{ab}$	69	$2,832^{b}$	70	2,845	73	3,173	45	<0.01	<0.01
Milk	$3,159^{a}$	68	$3,019^{ab}$	69	$2,825^{b}$	70	2,837	73	3,165	45	<0.01	<0.01
Calves (n = 127)[4]	11^{a}	0.2	9^{b}	0.2	8^{c}	0.2	9	0.2	9	0.1	<0.01	0.42
Costs[3]	$1,964^{a}$	64	$1,877^{ab}$	65	$1,729^{b}$	66	1,743	69	1,970	42	0.03	<0.01
Partially mixed ration	1,122	15	1,106	15	1,088	16	1,075	16	1,136	10	0.27	<0.01
Concentrate	$489^{a\dagger}$	15	$441^{a\dagger}$	15	387^{b}	15	403	16	475	10	<0.01	<0.01
Inseminations	69^{a}	6	63^{a}	6	41^{b}	6	56	6	59	4	<0.01	0.67
Veterinary treatments	95	(70-129)	88	(64-121)	74	(53-103)	55	(39-77)	132	(107-162)	0.50	<0.01
Culling due to health issues (n = 15)[5]	1,489	187	1,116	150	1,034	190	1,912	232	514	95	0.20	<0.01
Mean for all cows (n = 150)[6]	72	45	88	47	34	18	98	68	53	18	NA	NA
Not becoming pregnant (n = 14)[7]	452	97	181	83	329	43	405	87	236	41	0.09	0.08
Mean for all cows (n = 150)[6]	14	10	6	4	55	19	30	17	23	8	NA	NA
Net partial cash flow[3]	1,205	71	1,150	72	1,103	74	1,102	76	1,203	47	0.58	0.26
Net partial cash flow[8] – weighted (n = 150)	1,281	NA	1,205	NA	1,130	NA	1,171	NA	1,211	NA	NA	NA
Net partial cash flow[9] – weighted (N = 153)	1,276	NA	1,205	NA	1,108	NA	1,171	NA	1,198	NA	NA	NA

$^{a\text{-}c}$Different superscript indicates a difference among LSM from VWP or parity class ($P < 0.05$).

[1] Costs for veterinary treatments include medicine price, labor costs, and costs for discarded milk. *P*-values based on log transformation, data are back-transformed, and confidence interval (CI) is shown.

[2] VWP = voluntary waiting period. Par = parity (primiparous or multiparous) in the first lactation in the experiment. Interaction VWP × parity class was never significant and therefore not included in the models.

[3] Unweighted for the lactation length of an animal in the experiment.

[4] Revenues for calves; for cows that had a second calf in the experiment.

[5] LSM of culling costs for cows that were culled due to health issues in the experiment.

[6] Mean per group, also including cows that were not culled and that did become pregnant. No statistical analysis was performed, so *P*-values are not available.

[7] LSM of culling costs for cows that did not become pregnant in the experiment within 300 DIM, these cows left the experiment at approximately 530 days in lactation or earlier when production < 10L/d.

[8] Mean, weighted for the lactation length of an animal in the experiment.

[9] Mean, weighted for the lactation length of an animal in the experiment, including all cows.

[†] Similar symbol indicates a trend in difference among LSM from VWP or parity class ($P < 0.10$).

3.3 Cow factors associated with yearly net partial cash flow

Yearly net partial cash flow was first assessed for primiparous cows and multiparous cows separately. For primiparous cows, the first quartile of NPCF per cow per year was €1,037/yr and the third quartile of NPCF per cow per year was €1,364/yr (Figure 3a). For multiparous cows, the first quartile of NPCF per cow per year was €960/yr and the third quartile of NPCF per cow per year was €1,291/yr (Figure 3b).

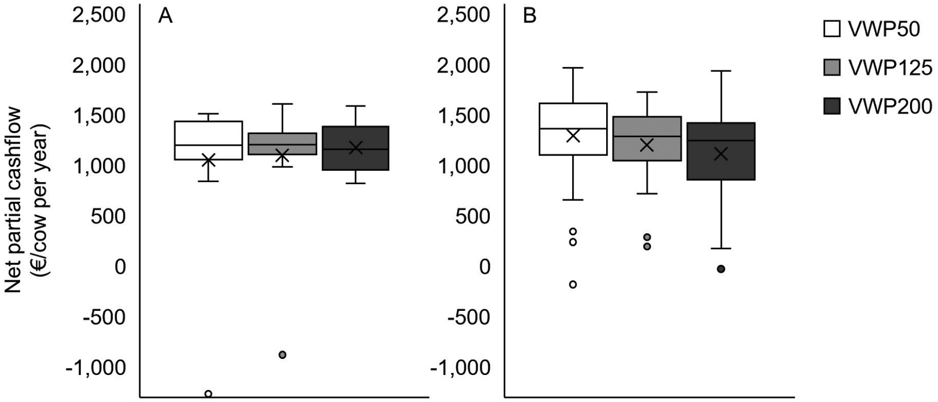


Figure 3. Net partial cash flow (€/cow per year) for primiparous (A) and multiparous (B) cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, VWP200) (n = 150). The × in the box indicates the mean, the solid black line in the box indicates the median, top and bottom of the box are the first and the third quartiles, and circles indicate the outliers.

Based on the yearly NPCF in the study, cows were divided in 3 EC. The VWP did not affect the chance to be in 1 of 3 EC (Table 4). The effects of EC, VWP, parity class, and all 2-way interactions on several cow characteristics were tested. The effect of EC on lactation persistency depended on VWP: cows in VWP125 tended to have greater lactation persistency in EC2 (−0.051 kg/d) compared with EC3 (−0.077 kg/d, *P* = 0.09). Within EC1, cows in VWP200 had greater lactation persistency compared with cows in VWP125 (−0.042 vs. −0.075 kg/d, *P* = 0.02). Within EC3, cows in VWP200 had greater lactation persistency (−0.048 kg/d) compared with cows in VWP50 (−0.076, *P* = 0.04) and tended to have greater lactation persistency compared with cows in VWP125 (−0.077, *P* = 0.07). Cows in EC3 (>€1,400/yr) had greater lactation yield (milk, protein, fat, or lactose) per ED (kg/d) and fewer veterinary treatments in the experiment compared with cows in EC1 (<€1,100/yr; Table 5). Moreover, the effect of EC on lactose content depended on parity class: primiparous cows had greater lactose content in

EC1 (4.4%) compared with EC2 (4.1%, *P* = 0.02) or compared with EC3 (4.1%, *P* = 0.03), whereas for multiparous cows lactose content did not differ among EC. Both CFSI and CInt were not different among the 3 EC.

Table 4. Number of cows from each voluntary waiting period group (50 d, 125 d, or 200 d: VWP50, VWP125, or VWP200) in each economic class (EC1: <1,100 €/year, EC2: 1,100-1,400 €/year, EC3 >1,400 €/year). Proportion of cows from each VWP in the 3 EC between brackets (%)

	VWP50	VWP125	VWP200	Total
EC1	14 (27)	15 (31)	21 (43)	50
EC2	18 (35)	18 (37)	15 (31)	51
EC3	20 (38)	16 (33)	13 (27)	49
Total	52 (100)	49 (100)	49 (100)	150

Chi-square = 3.46, *P*-value = 0.48.

When the effect of cow characteristics in the first 6 wk after the first calving within the experiment on yearly NPCF was evaluated, the final multivariate model included VWP class and parity class as class variables, and MY, FPCM yield, BW, the interaction BW × VWP, maximum yield, the interaction maximum yield × VWP, and the breeding value for persistency as continuous variables (Table 6). In this model, mean MY in the first 6 wk was negatively associated with yearly NPCF, whereas mean FPCM yield in the first 6 wk was positively associated with yearly NPCF. Mean BW in the first 6 wk was positively associated with yearly NPCF, mostly in VWP200, and mostly for primiparous cows. Maximum yield in the first 6 wk was positively associated with yearly NPCF, mostly in VWP50. Moreover, the breeding value for persistency was positively associated with yearly NPCF.

Table 5. Cow characteristics for cows in different economic classes (EC1: <1,100 €/cow per year, EC2: 1,100-1,400 €/cow per year, EC3 >1,400 €/cow per year) that were not culled during the experiment (n = 150)

	EC1		EC2		EC3		*P*-value[1]		
	LSM	SEM (CI)	LSM	SEM (CI)	LSM	SEM (CI)	EC	VWP	Par
n cows	50		51		49				
CFSI[2] (d)	140	2	138	1.7	140	1.9	0.67	<0.01	0.89
Calving interval (d)	452	8	444	7	435	7	0.26	<0.01	0.21
Lactation persistency[3] (kg/d)[*]	-0.061	0.005	-0.061	0.004	-0.067	0.004	0.50	<0.01	<0.01
Milk yield per ED[4] (kg/d)	$23.3^{b\dagger}$	0.6	$25.2^{b\dagger}$	0.6	28.2^{a}	0.6	<0.01	<0.01	<0.01
Protein yield per ED (kg/d)[**]	0.84^{c}	0.02	0.93^{b}	0.02	1.04^{a}	0.02	<0.01	0.16	<0.01
Fat yield per ED (kg/d)[**]	1.02^{c}	0.02	1.12^{b}	0.02	1.27^{a}	0.02	<0.01	0.89	<0.01
Lactose yield per ED (kg/d)	$1.05^{b\dagger}$	0.03	$1.14^{b\dagger}$	0.03	1.27^{a}	0.03	<0.01	<0.01	<0.01
Protein content[5] (%)	3.5	0.04	3.5	0.04	3.4	0.04	0.66	<0.01	0.58
Fat content[5] (%)	4.2	0.08	4.2	0.08	4.2	0.08	0.79	<0.01	0.09
Lactose content[5] (%)[***]	4.2^{a}	0.04	4.1^{b}	0.03	4.1^{b}	0.04	0.01	0.15	<0.01
Veterinary treatments[6]	7.8^{a}	(6.1-10)	4.3^{b}	(3.4-5.4)	2.9^{c}	(2.3-3.7)	<0.01	0.50	<0.01
Body weight (kg)	657	8	660	8	665	8	0.79	0.63	<0.01
Body condition score	2.7	0.06	$2.8^{\dagger}$	0.06	$2.6^{\dagger}$	0.06	0.09	<0.01	0.29
Net partial cash flow (€/cow per year)[**]	658^{c}	44	$1{,}255^{b}$	44	$1{,}598^{a}$	46	<0.01	0.20	0.43

[a-c] Different superscript indicates a difference among LSM from VWP or parity class ($P < 0.05$).

[*] Interaction EC × VWP in the model: $P = 0.04$.

[**] Interaction VWP × parity class in the model: protein yield: $P = 0.03$; fat yield: $P = 0.01$; net partial cash flow: $P = 0.04$.

[***] Interaction EC × parity class in the model: $P = 0.02$.

[1] EC = economic class. VWP = voluntary waiting period. Par = parity (primiparous or multiparous) in the first lactation in the experiment.

[2] CFSI = calving to first service interval.

[3] Lactation persistency between day 100 and start dry-off.

[4] ED = experimental day: yield per day in the experiment.

[5] Mean content during the experiment.

[6] Number of veterinary treatments in the experiment. *P*-values based on log transformation, data are back-transformed, and confidence interval (CI) is shown.

[†] Similar symbol indicates a trend in difference among LSM within the row ($P < 0.10$).

Table 6. Final multivariable model for prediction of net partial cash flow (€/cow per year) for cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, or VWP200) (n = 150) (LSM ± SEM or regression coefficient (β) with standard error and range). The final multivariate model was based on 13 univariate models, with individual early lactation variables as independent variable, to identify potential predictors for net partial cash flow after different VWP

Variable	Category	LSM (SEM) or *β (SE)*	*Range*[1]	*P*-value
VWP	50	1,572 (114)		0.23
	125	1,447 (117)		
	200	1,425 (115)		
Parity	Primiparous	1,821 (207)		0.06
	Multiparous	1,142 (42)		
Milk yield[2]		*-77 (27)*	*17-52*	<0.01
FPCM yield[2]		*62 (17)*	*19-56*	<0.01
Body weight[2]		*1.8 (2.2)*	*493-870*	0.16
Body weight × VWP	50[3]	*0 (-)*	*520-836*	0.02
	125	*1.8 (1.3)*	*493-825*	
	200	*4.0 (1.4)*	*512-870*	
Body weight × parity class	Primiparous[3]	*0 (-)*	*493-646*	0.03
	Multiparous	*-4.4 (2.1)*	*526-870*	
Maximum yield[2]		*65 (21)*	*21-60*	0.04
Maximum yield × VWP	50[3]	*0 (-)*	*22-60*	<0.01
	125	*-31 (12)*	*24-59*	
	200	*-45 (13)*	*21-58*	
Breeding value persistency		*32*	*92-113*	<0.01

[1] Range for milk yield FPCM (fat- and protein-corrected milk) yield, and maximum yield in kg/day; range for body weight in kg.
[2] Measured in the first 6 wk after the first calving within the experiment.
[3] Reference category.

3.4 Sensitivity analysis

When prices for fat, protein, and lactose were minimal or maximal, yearly NPCF of cows in VWP50 was more affected compared with yearly NPCF of cows in VWP200 (Table 7). A change in feed or calf prices, or a change in labor costs for calving cows or for inseminations also had a greater effect on yearly NPCF of cows in VWP50 compared with cows in VWP200. The VWP did not affect the change in yearly NPCF when labor costs for veterinary treatments were minimal or maximal.

When prices for fat, protein, and lactose were minimal or maximal, yearly NPCF of multiparous cows was more affected compared with yearly NPCF of primiparous cows. Moreover, a change in feed prices or labor costs for veterinary treatments had a greater effect on yearly NPCF of multiparous cows compared with primiparous cows. Parity class did not affect the change in yearly NPCF when calf prices were minimal or maximal, or when labor costs for calving cows or inseminations were minimal or maximal.

Table 7. Relative change of net partial cash flow (NPCF; €/cow per year) for minimum and maximum price of milk, feed, and calves, and for minimum and maximum labor costs around calvings (calving cow labor), inseminations (insemination labor), and veterinary treatments (veterinary treatment labor) for cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, VWP200) (n = 150)

	Voluntary waiting period						Parity class				*P*-value[1]	
	VWP50		VWP125		VWP200		Primiparous cows		Multiparous cows		VWP	Par
	LSM	SEM (CI)	LSM	SEM (CI)	LSM	SEM (CI)	LSM	SEM (CI)	LSM	SEM (CI)		
NPCF	1,205	71	1,150	72	1,103	74	1,102	76	1,203	47	0.58	0.26
Milk price – min	-880^{b}	18	-844^{ab}	18	-793^{a}	19	-795	19	-882	12	<0.01	<0.01
Milk price – max	+808^{a}	17	+775^{ab}	17	+728^{b}	17	+730	18	+810	11	<0.01	<0.01
Feed price – min	+237^{a}	4.1	+225^{ab†}	4.2	+212^{b†}	4.3	+214	4.5	+236	2.7	<0.01	<0.01
Feed price - max	-21^{b}	0.3	-20$^{a\dagger b}$	0.3	-19^{a†}	0.4	-19	0.4	-21	0.2	<0.01	<0.01
Calf price – min	-58^{b}	2.9	-46^{a}	2.9	-38^{a}	3.0	-49	3.1	-46	1.9	<0.01	0.40
Calf price – max	+58^{a}	2.9	+46^{b}	2.9	+38^{b}	3.0	+49	3.1	+46	1.9	<0.01	0.40
Calving cow labor – min	+31^{a}	1.5	+25^{b}	1.6	+20^{b}	1.6	+26	1.7	+24	1.0	<0.01	0.40
Calving cow labor – max	-57^{b}	2.9	-46^{a}	2.9	-37^{a}	3.0	-48	3.1	-45	1.9	<0.01	0.40
Insemination labor – min[2]	+25^{a}	(21-30)	+22^{a}	(19-26)	+15^{b}	(13-18)	+20	(16-24)	+21	(19-24)	<0.01	0.47
Insemination labor – max[2]	-25^{b}	(21-30)	-22^{b}	(19-26)	-15^{a}	(13-18)	-20	(16-24)	-21	(19-24)	<0.01	0.47
Veterinary treatment labor – min[2]	+33	(24-46)	+35	(25-50)	+27	(19-38)	+19	(13-27)	+52	(41-65)	0.49	<0.01
Veterinary treatment labor – max[2]	-94	(73-120)	-87	(68-113)	-71	(55-93)	-61	(46-80)	-115	(97-136)	0.27	<0.01

[a,b] Different superscript indicates a difference among LSM from VWP ($P < 0.05$).

[1] VWP = voluntary waiting period. Par = parity (primiparous or multiparous) in the first lactation in the experiment. Interaction VWP × parity class was never significant and therefore not included in the models.

[2] *P*-values based on log transformation, data are back-transformed, and confidence interval (CI) is shown.

[†] Similar symbol indicates a trend in difference among LSM from VWP ($P < 0.10$).

4 Discussion

In this study, when VWP was extended until 125 or 200 d, cows had a greater CSFI, and a greater CInt. Moreover, over the complete experimental period, production of milk, protein, fat, and lactose was greater after longer VWP, as cows with a longer VWP had more lactating days during the experiment. Other studies also reported greater total lactation yields of milk, protein, fat, and lactose for cows with extended VWP or CInt, attributed to the greater lactation length of these cows (Van Amburgh et al., 1997; Rehn et al., 2000; Österman and Bertilsson, 2003). Cows with an extended VWP of 125 or 200 d had greater PMR supply in the experiment compared with a VWP of 50 d. An extended VWP, however, did not result in greater concentrate supply during the experiment. Therefore, cows with a VWP of 200 d received less concentrate per year compared with cows in VWP50. Similarly, number of inseminations or number of veterinary treatments during the complete experiment was not affected by VWP.

Moreover, probability of being culled due to health issues in the experiment was not affected by VWP. Numerically, more cows in VWP125 (7) and VWP200 (7) were culled during the experiment compared with VWP50 (4), possibly related to having more days in the experiment. From these culled cows in VWP125 and VWP200, 3 cows from each group were culled in the first 6 wk of the second lactation within the experiment. This could possibly be related to the increased BCS of cows with a longer VWP at the end of lactation and in the beginning of the next lactation (Burgers et al., 2021b). However, the sample size in this study was too small to draw conclusions about culling rate in the different VWP groups. In an earlier study with 2,711 cows, it was observed that multiparous cows with a waiting period of 88 d tended to have greater replacement costs within 1 lactation compared with multiparous cows with a waiting period of 60 d (Stangaferro et al., 2018).

The probability to not become pregnant within 300 DIM in the experiment tended to be affected by VWP. Although the 3 VWP groups did not statistically differ, numerically more cows in VWP200 did not conceive within the experiment compared with the other 2 VWP groups. In this study, all cows had until 300 DIM to conceive. As a result of this approach, cows with a longer VWP had less time to become pregnant. Moreover, 4 cows from VWP200 were never inseminated, from which 3 were culled before the end of the VWP or shortly after (219 d). Although 9 cows from VWP200 did not conceive in this study, the cows in this group had more normal ovarian cycles around the end of the VWP, and fewer days until pregnancy after the end of the VWP (Ma et al., 2020).

As an alternative to calculating output as the sum for the complete lactation, variables could be expressed per year to allow for the comparison of results of cows with different VWP and therefore different lactation lengths. Hence, in this study, revenues and costs were calculated for each VWP group per cow per year. Yearly revenues were lower in VWP200 compared with VWP50, mainly due to lower yearly revenues from milk. Earlier modeling studies also found a reduction in milk production or milk revenues for most cows when VWP was extended (Groenendaal et al., 2004; Steeneveld and Hogeveen, 2012; Kok et al., 2019). For example, when CInt was extended with 2 or 4 mo for both primiparous and multiparous cows, milk production was reduced by 4% or 7% (Kok et al., 2019). When another study included only high-producing cows, however, income from milk increased when VWP was extended from 90 to 150 d for primiparous cows or from 60 to 120 d for multiparous cows (Arbel et al., 2001). In practice, farmers that want to extend the CInt on their farm select specific cows for an extended VWP. For example, when farmers deliberately extended the CInt for (part of) their herd, cows with the greatest 305-d production had longer CInt (Burgers et al., 2021a). Therefore, in those cases, the losses in milk revenues might be limited. Moreover, minimum or maximum price for milk solids had greater effect on the yearly NPCF of cows in VWP50 compared with cows in VWP200. Probably the reason is the greater MY for cows in VWP50, resulting in a relatively greater effect of fluctuation in milk price on the cash flow compared with cows in VWP200. Also in an earlier study, when costs for a lower milk production were lower, a delayed insemination had less negative effect on the yearly economic result of cows compared with when costs for a lower milk production were higher (Steeneveld and Hogeveen, 2012). Therefore, in the current study, when prices for milk solids were low, the yearly NPCF of cows with a VWP of 200 d approached the yearly NPCF of cows with a VWP of 50 d (€310 vs. 325). Next to the lower milk revenues, net returns from calves were lower when VWP was longer. In the current study, we assumed that the ratio of male to female calves born was 50% for all 3 groups. This assumption might not reflect the actual sex of calves born. Moreover, we did not distinguish between the value of heifers that were kept for replacement or the value of calves that were sold at 2 wk of age. As calf revenues did not have a large contribution to the net partial cash flow, the effect of these assumptions on the economic result is expected to be limited. Minimum or maximum prices for calves, or minimum or maximum costs for labor associated with calving cows, had greater effect on the yearly NPCF of cows in VWP50 compared with cows in VWP200, as more calves were born per year in this group.

In the current study, the lower revenues for cows in VWP200 were partly compensated by lower costs for cows in VWP200 compared with VWP50, mainly due to lower costs for concentrate supply. These lower costs for concentrate in longer VWP could be explained by more days with lower milk production per year and fewer days in peak yield per year (Dekkers et al., 1998; Lehmann et al., 2014; Kok et al., 2019). As more concentrate was supplied per year to cows in VWP50, minimum or maximum feed prices had greater effect on the yearly NPCF of cows in VWP50 compared with cows in VWP200. Next to the lower yearly feed costs, yearly insemination costs were lower for cows in VWP200. These lower costs for insemination could be explained by better fertility of cows in VWP200 (Ma et al., 2020) and better fertility of cows with a longer VWP in general (Niozas et al., 2019b). The number of inseminations in the complete experiment did not significantly differ among the VWP groups, but more days in lactation with an equal number of inseminations per lactation leads to lower costs for inseminations per year. As cows in VWP50 had more inseminations per year, minimum or maximum costs for labor around inseminations had a greater effect on the yearly NPCF of cows in VWP50 compared with cows in VWP200. The mean yearly costs for culling due to health issues were €38 greater in VWP50 compared with VWP200, possibly related to similar culling within one lactation, so costs become greater for cows with shorter lactation lengths than for cows with longer lactation lengths when culling is expressed per year. The mean yearly costs for not becoming pregnant were €41 greater in VWP200 compared with VWP50, possibly related to their shorter time to become pregnant. Costs for veterinary treatments did not differ among the 3 VWP groups. The costs for culling and veterinary treatments, together with the insemination costs, also had a relatively small contribution to the yearly NPCF. Moreover, the VWP did not affect the change in yearly NPCF as a result of minimal or maximal costs for veterinary treatments, possibly associated with a low disease incidence and the relatively low number of cows for assessment of treatments. In addition, this study included only cows that had a SCC below 250,000 before the previous dry-off and that were expected to complete a full lactation. In theory this could have resulted in a selection bias, as only relatively healthy cows were included.

Due to the partial compensation of the lower revenues by lower costs after longer VWP, the difference in yearly NPCF among the VWP groups was reduced, and VWP was not significantly associated with the yearly NPCF. The sample size of the current study was, however, relatively small for an economic analysis including costs of culling. Yearly NPCF was, on average, €55/cow per year lower when cows had a VWP of 125 compared with a VWP of 50 d, and was €102/cow per year lower when cows had a VWP of 200 d compared with a VWP of 50 d. Although these differences were not statistically significant in this study, they could be relevant for farmers in practice. When VWP would be extended up to 200 d for all cows on a farm, the implications for the economic result could be large on a herd level if all these cows would realize lower cash flows. However, it can be expected that not all cows with a VWP of 200 d have €102 lower yearly cash flows, as some cows might be better suited for a longer VWP than others (Lehmann et al., 2017; Kok et al., 2019; Burgers et al., 2021b). This was also shown by the large variation in yearly NPCF among individual cows. Selecting the more suitable cows for a longer CInt might limit the reductions in NPCF on a herd level. To account for the lactation length of individual cows in the experiment, we calculated the weighted yearly NPCF per VWP group. As such, cows with fewer days in the study had a smaller contribution to the average NPCF than cows with more days in the study. The differences in NPCF among the 3 VWP were greater in the weighted averages compared with the unweighted NPCF. As the weighted yearly NPCF accounted for the lactation length of a cow, the 3 cows that were culled before 70 DIM and therefore had extremely high yearly costs for culling could be included. The difference between the weighted NPCF excluding these 3 cows and the weighted NPCF with all cows was small, and it can be concluded that these 3 cows made a limited contribution to the results of the experiment.

One of the reasons that the yearly NPCF among the 3 VWP groups did not significantly differ was probably the large variation in NPCF among individual cows. To evaluate why some cows performed better in terms of yearly NPCF than other cows, cows were divided into 3 EC based on their yearly NPCF. The effect of these EC on several cow characteristics was studied. The effect of EC on lactation persistency depended on VWP, where, only for cows in VWP125, the lactation persistency was greater in EC2 (€1,100–1,400/yr) compared with EC3 (>€1,400/yr). In contrast, earlier studies found an increased NPCF in extended lactations when lactation persistency was increased (Kok et al., 2019). Possibly, in the current study, the improved lactation persistency was related to a lower peak production (Dekkers et al., 1998), which is also an important factor for the economic results of cows. Lactation persistency, however, could be important for maintaining a healthy BCS at the end of an extended lactation. When BCS is increased in late lactation as a result of an extended VWP, this could result in an increased risk for diseases after the next calving (Roche et al., 2009).

In the current study, the interaction between VWP and parity class never affected the yearly revenues and costs. In other studies, when CInt was extended, results for MY, milk revenues, or NPCF were often different for primiparous cows than for multiparous cows (Österman and Bertilsson, 2003; Lehmann et al., 2016; Kok et al., 2019). When we forced the interaction between VWP and parity class in the model in the current study, numerically the yearly NPCF for multiparous cows decreased with longer VWP (€1,294 vs. 1,200 vs. 1,114 for VWP50, VWP125, and VWP200), but the yearly NPCF for primiparous cows did not decrease with longer VWP (€1,050 vs. 1,099 vs. 1,175 for VWP50, VWP125, and VWP200). Similarly, cash flow of primiparous cows numerically increased when VWP was extended from 60 d to 88 d, and cash flow of multiparous cows numerically decreased when VWP was extended (Stangaferro et al., 2018). This could be related to the greater lactation persistency of primiparous cows compared with multiparous cows (Lehmann et al., 2017; Kok et al., 2019; Burgers et al., 2021b), as for primiparous cows, revenues from milk also did not decrease when VWP was extended (€2,857 vs. 2,809 vs. 2,879 for VWP50, VWP125, and VWP200).

Other cow characteristics than parity could also play a role in the lactation performance of cows. Yearly NPCF of cows with different VWP could be predicted by the maximum yield and the mean BW in the first 6 wk after the first calving in the experiment. In the model, maximum yield in the first 6 wk was positively related to NPCF per cow per year for cows in all 3 VWP, but less for cows in VWP125 and VWP200 compared with cows in VWP50. Possibly this could be explained by a reduced lactation persistency related to a greater peak yield (Dekkers et al., 1998). Moreover, when VWP and thus CInt is shorter, early lactation takes up a relatively greater part of the complete lactation; production during this period is therefore more important than when VWP, and thus CInt is longer. Mean BW in the first 6 wk was positively related to NPCF per cow per year for cows in all VWP, but mostly for cows in VWP200. It can be hypothesized that BW in early lactation could play a role in persistency later in lactation (Dekkers et al., 1998). Especially after longer VWP, persistency is an important factor to maintain MY in these extended lactations.

In the current study, cows were randomly divided over 3 different VWP. Moreover, the ration of the cows in the 3 VWP groups was the same. In practice, farmers often select specific cows for an extended lactation, based on MY level, body condition, or a combination of factors (Burgers et al., 2021a). Moreover, some farmers have deliberate final lactations, where cows are not inseminated but remain on the farm and are milked. In the current study, in total 3 primiparous cows and 11 multiparous cows left the experiment because they did not conceive within 100 d after the end of the VWP. These cows were mostly able to produce sufficient amounts of milk at the end of their lactation and stay in the experiment for a long time, as illustrated in Figure 4. Of these cows, 4 multiparous cows were culled because their production dropped below 10 L/d before 530 DIM, between 393 and 495 DIM. Possibly, 10 L/d is a low cut-off value in practice and cows are culled earlier on commercial dairy farms, as these production levels could be insufficient for a productive dairy farm. If, in this study, cows had been culled earlier in lactation due to a higher cut-off value, culling costs of these cows could be higher due to lower culling age. If, for example, the cut-off value had been 15 L/d, these 4 cows would have been culled 103 d earlier on average. This effect, however, can be expected to be limited because, on average, culling age of these cows would in that case be reduced with only ± 5%, or with ± 8% if the cut-off value had been 20 L/d. In addition, most cows were able to remain in the study until 530 DIM because they had adequate production levels (i.e., on average 16 kg/d in the final 7 d in the study). This study stopped following these other cows at approximately 530 DIM; however, in practice these cows might stay at the farm for an even longer period as long as they produce sufficient milk.

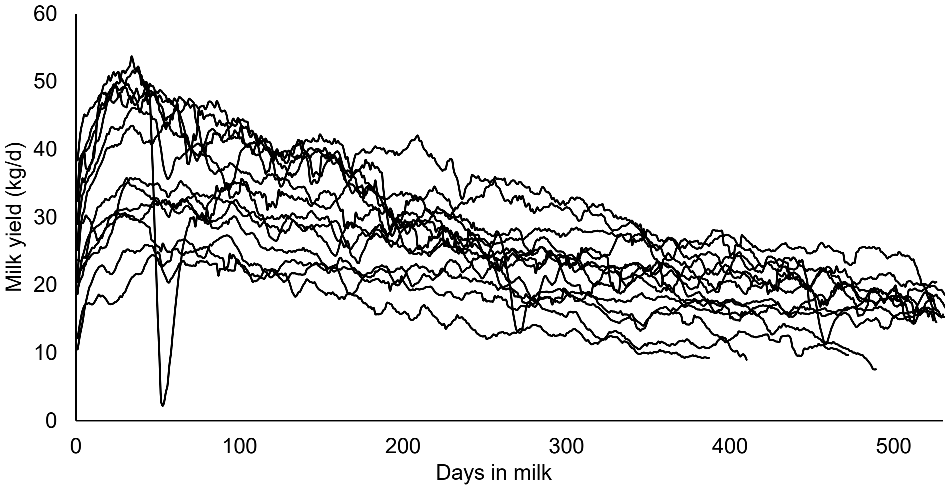


Figure 4. Milk yield based on the weekly rolling average of cows that did not conceive in the experiment and left the experiment at 530 DIM or earlier when their milk yield dropped below 10 L/d (n = 14).

5 Conclusions

In an experiment where cows were randomly assigned to different VWP and managed accordingly, cows with a VWP of 50 d had greater total yearly revenues and greater total yearly costs compared with cows with a waiting period of 200 d. Total revenues and costs per year for cows in VWP125 were similar to those of cows in VWP50. The yearly NPCF was not affected by the VWP. Milk revenues and feed costs contributed the most to the yearly NPCF. Cows with a greater yearly NPCF had greater production of milk, protein, fat, and lactose, and a lower number of veterinary treatments. Neither VWP nor CInt were different for cows with greater yearly NPCF compared with cows with lower yearly NPCF. For cows in VWP50, a greater maximum yield in the first 6 wk was more strongly associated with a greater yearly NPCF than for cows after longer VWP.

6 Acknowledgments

This study was financed by DairyNL (ZuivelNL; organization of the Dutch dairy supply chain, The Hague, the Netherlands) and the Dutch Ministry of Agriculture, Nature and Food Quality (LNV, The Hague) as part of the research program One Health for Food (1H4F, The Hague). The authors thank the staff of the Dairy Campus (Leeuwarden, the Netherlands) and veterinarian Betsie Krattley of University Livestock Practice (Harmelen, the Netherlands) for information on the costs of treatments and related labor. The authors have not stated any conflicts of interests.

Appendix

Table A1. Treatments, duration of treatments, price per complete treatment, labor costs, and waiting time for milk per treatment for all medication used in the experiment

Treatment	Treatment duration	Price (€/complete treatment)	Labor costs (€/complete treatment)	Waiting time (d)
Acegon	1 treatment, 1 d	8	15	0
Albipen	3 treatments, 3 d	26	30	6
Avuloxil	3 treatments, 1.5 d	9.5	30	4
Biodyl	1 treatment, 1 d	11.5	15	0
Borgal	3 treatments, 3 d	13	30	3
Bovi-C3	1 treatment, 1 d	6.4	15	0
Bovical	2 boluses	12	15	0
Buscopan	1 treatment, 1 d	15	15	7
Cai pan	1 treatment, 1 d	2.9	5	0
Calci TAD 25	1 treatment, 1 d	5	15	0
CA-MG IV	1 treatment, 1 d	5.69	15	0
Diatrim 24	3 treatments, 3 d	22.5	30	2
Dinolytic	1 treatment, 1 d	5	15	0
Dofatrim	3 treatments, 3 d	22.5	30	3
Drench	1 treatment, 1 d	14	30	0
Engemycine 10	3 treatments, 3 d	13.2	30	5
E-pil	1 bolus	6.5	15	0
Fyto-stop powder	1 treatment, 1 d	6.35	15	0
Glucamagnesium	1 treatment, 1 d	15	15	0
Glucose 30	1 treatment, 1 d	10.88	15	0
Mamyzin	3 treatments, 3 d	30	30	4
Placenta capsule	1 treatment, 1 d	4.7	15	4
Noroseal	1 treatment (4 teats)	8	5	0
Novem	1 treatment	12.5	15	5
Orbenin dry	1 treatment (4 teats)	6	5	42
Orbenin extra dry	1 treatment (4 teats)	8.84	5	42
Orbenin lactation	3 treatments, 6 d	13.92	30	4
Orbeseal	1 treatment (4 teats)	8	5	0
P-pil	2 boluses	7	15	0
Pen & strep	3 treatments, 3 d	14	30	5
Prid	1 treatment, 1 d	26.25	15	0
Procapen	3 treatments, 3 d	15	30	6
Propylene glycol	2 doses	10	15	0
Revozyn	3 treatments, 3 d	45.78	30	4
Rimadyl	1 treatment, 1 d	14.54	15	0
Rotavec Corona	1 treatment, 1 d	16.49	5	0
Rumiactif	1 treatment, 1 d	6.4	15	0
TMP/SMZ	3 treatments, 3 d	22.5	30	4
Ubrolexin	2 treatments, 2 d	7.11	30	5
Ubropen	3 treatments, 3 d	14.63	30	6
Ubroseal	1 treatment (4 teats)	8	5	0

Chapter 6

General discussion

1 Introduction

High-producing dairy cows usually experience a negative energy balance (NEB) in the first 3 months after calving, due to a steep increase in milk production and a more limited increase in feed intake during that time (Butler et al., 1981; Rastani et al., 2005). The calving process and the NEB are associated with an increased risk for diseases (Ingvartsen et al., 2003; Friggens et al., 2004). In most dairy systems, farmers aim for their cows to have a calf every year, resulting in a yearly calving moment, a yearly peak in milk yield and, as a trade-off, a yearly high-risk period for diseases. Extending the calving interval (CInt) by extending the voluntary waiting period for insemination (VWP) would reduce the frequency of calving moments per cow. This is expected to reduce the disease frequency associated with the transitions around calving. With fewer calving moments, however, cows also have fewer days in peak milk production. Moreover, cows spend more time in late lactation, where milk production is usually lower. This may not only decrease the milk production per lactation or per day, but it also could cause fattening of cows at the end of the lactation, what in turn may increase the risk for metabolic problems after the subsequent calving (Gillund et al., 2001; Roche and Berry, 2006; Niozas et al., 2019a).

The work described in chapters 2, 3, 4, and 5 in this thesis was carried out with as aim to evaluate the consequences of an extended lactation for milk production, reproductive performance, health, metabolism, and economic performance. In this chapter, first I will discuss the difference between an extended CInt, an extended calving to first service interval (CFSI), and an extended VWP. Second, I will discuss the consequences of extended lactations for milk production, reproductive performance, cow health and metabolism, and economic performance, where results of this thesis are compared with earlier modeling, observational, and experimental studies. Next, I will discuss cow characteristics that could be related with lactation performance of cows with different lactation lengths. Finally, I will discuss the possibilities and implications of applying extended lactations in practice, and the practical and scientific knowledge gaps.

2 Calving interval, calving to first service interval, and voluntary waiting period

In the debate about optimal reproductive decision making, it is important to distinguish 3 terms that are used by studies that investigate extended lactations: extended calving interval (CInt), extended calving to first service interval (CFSI), and extended voluntary waiting period (VWP) (Figure 1). The VWP is a decision made beforehand, and part of the management strategy of farmers. After the VWP, at first detected estrus the first artificial insemination (AI) will take place, resulting in the CFSI. The CFSI is a direct consequence of the VWP but is also influenced by the quality of estrus detection. An extended CFSI may be the result of cows not showing estrus earlier, failing to detect estrus, or cows suffering from diseases in early lactation which made the farmer decide to wait with insemination. The CInt of a cow is the result of the VWP, the quality of estrus detection and the conception of cows. Conception is associated with the number of inseminations it takes for a cow to get pregnant. As such, an extended CInt may be the result of failed estrus detection or failed inseminations. The CInt is only available for cows that have a subsequent calf, and consequently, the CInt does not give complete information for the entire herd. Especially cows with bad reproductive performance may not become pregnant and may lack a CInt.

Farmer's decisions, management, and cow health influence the VWP, CFSI, and CInt. Some of the factors that affect the VWP, CFSI, and CInt are direct decisions of a farmer: the VWP and the culling policy. The decision to continue with inseminating until late in lactation might increase the CInt, whereas the decision to cull a cow after a specific number of unsuccessful inseminations might decrease the CInt. Other factors have an indirect effect on the VWP, CFSI, and CInt: the health and fertility of the cow, and the quality of estrus detection. Impaired health and fertility may imply a low conception rate, which could increase the CInt. Moreover, a poor detection of estrus may increase both the CFSI and the CInt.

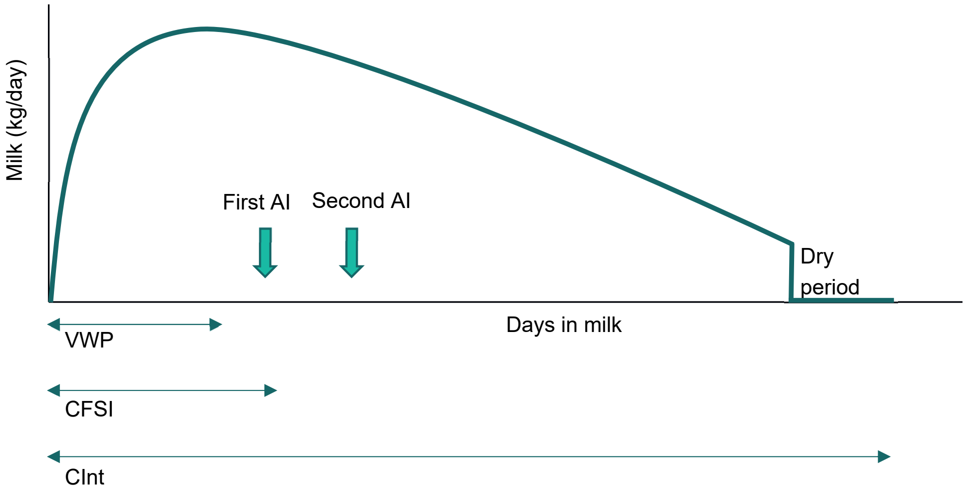


Figure 1. Schematic presentation of the lactation cycle of a dairy cow with a voluntary waiting period (VWP), a calving to first service interval (CFSI), and a calving interval (CInt). In this schematic presentation, the second AI (artificial insemination) was the successful insemination, resulting in the CInt.

Modeling studies, observational studies, experimental studies, and combinations of these have been used to investigate extended lactations. In modeling studies, input is based on retrospective farm data or effect studies. In observational studies, retrospective farm data is analyzed, where consequences of an extended CFSI or CInt in practice can be investigated. The disadvantage of this approach is that it is unknown if an extended CInt is the result of impaired fertility, diseases in early lactation, or a deliberate decision of the farmer. The advantage of this approach is that usually large datasets are available over long periods of time, which increases the power of these studies. In experimental studies, cows are randomly distributed over different lengths of VWP under controlled circumstances, and blocked for specific cow characteristics, to achieve an equal distribution of similar cows over different VWP. Moreover, differences in management among farms does not have an effect in experimental studies. As such, limited confounding effects of other factors may affect the results. Experimental studies, however, have limitations in terms of animal numbers, and are intensive and expensive. Therefore, large experimental studies on extended lactations are quite unique, as cows need to be followed for a long time under experimental conditions. Especially in order to study events that do not frequently occur, larger, observational, datasets are important, for instance when studying diseases, culling, or lifespan.

Therefore, in this thesis, 2 approaches were used to study the consequences of an extended lactation. First, an animal experiment was performed where 154 cows were blocked for parity, expected milk production, and the breeding value for persistency, and withing blocks cows were randomly assigned to a VWP of 50, 125 or 200 days. This experiment was performed at a research dairy farm, where cows could be intensively followed with multiple measurements considering milk production characteristics, body condition, feed intake, blood metabolites and hormones, and health. In addition, data of a network of commercial dairy farmers who deliberately extended the lactation for (part of) the herd was collected and analyzed. As these farms had deliberately extended lactations, management may be expected to be adjusted to these lactation lengths. For example, these farmers use specific feeding strategies, and breed for cows that are more suitable for extended lactations.

3 Milk production and lactation persistency

An extended lactation may affect the milk production by 1. fewer days in peak production relative to the complete lactation 2. more days in late lactation with a lower production relative to the complete lactation and 3. fewer days dry relative to the lactating days. To be able to compare milk production of cows with different lactation lengths, in most recent studies on extended VWP, milk production is expressed per day of CInt, also called milk production per feeding day (Lehmann et al., 2016) or effective lactation yield (Chapter 2; Kok et al., 2019).

Table 1 shows the consequences of different VWP in experimental studies for the milk production per day of CInt. Most studies presented their results separately for primiparous cows and multiparous cows. In our experiment (Chapter 3), for primiparous cows the VWP could be extended until 200 days with no effect on the milk production or the fat- and protein-corrected milk (FPCM) production per day of CInt. For multiparous cows, the VWP could be extended until 125 days with no effect on the milk or FPCM production per day of CInt. When the VWP was extended until 200 days for multiparous cows, however, milk or FPCM production per day of CInt did decrease with approximately 2.5 kg per cow per day after correction for the milk production during the first 6 weeks in lactation (Chapter 3). The VWP did not affect the production of fat, protein, or lactose per day of CInt (Chapter 3; Rehn et al., 2000). This may be explained by the greater fat and protein content and the lower milk production at the end of the lactation when the VWP was extended (Chapter 3, Silvestre et al., 2009). In another study, primiparous cows with a VWP of 150 days had a greater value-corrected milk production per day of CInt compared with primiparous cows with a VWP of 90 days (Arbel et al., 2001). For multiparous cows, the value-corrected milk production per day of CInt did not differ when the VWP was extended from 60 to 120 days (Arbel et al., 2001). In addition, multiparous cows with a VWP of 140 days had numerically a lower milk production compared with multiparous cows with a VWP of 50 days, while primiparous cows with a VWP of 140 days had numerically a greater milk production compared with primiparous cows with a VWP of 50 days (Rehn et al., 2000). The contrast between primiparous cows and multiparous cows in terms of milk production per day of CInt after an extension of the VWP can be explained by the difference lactation persistency between primiparous and multiparous cows.

Table 1. Milk production per day of calving interval for primiparous cows and multiparous cows with an extended voluntary waiting period in experimental settings

	Breed[1]	Parity	VWP	Milk production per day of calving interval
Burgers et al., 2021b (chapter 3)[2]	HF	Primiparous	50	22.9
			125	22.9
			200	24.6
		Multiparous	50	28.0[a]
			125	27.4[ab]
			200	25.4[b]
Niozas et al., 2019[3]	HF	All	40	30.6
			120	30.8
			180	30.5
Stangaferro et al., 2018[4]	H	Primiparous	60	27.8[a]
			88	29.3[b]
		Multiparous	60	35.3
			88	35.3
Österman and Bertilsson, 2003[5]	SRB	Primiparous	50	22.6
			230	23.9
		Multiparous	50	23.8
			230	22.8
Arbel et al., 2001	IH	Primiparous	90	26.4[a]
			150	27.0[b]
		Multiparous	60	31.4
			120	31.0
Rehn et al., 2000	SRB	Primiparous	50	18.1
			140	18.3
		Multiparous	50	20.0
			140	19.9
	SLB	Primiparous	50	21.0
			140	21.4
		Multiparous	50	25.3
			140	24.2
Van Amburgh et al., 1997[6]	HF	All	60	28.8
			150	31.0

[a,b] Different letters indicate a difference among means within a study and within a parity class.

[1] HF = Holstein Friesian; H = Holstein; SRB = Swedish Red and White; SLB = Swedish Holstein; IH = Israeli Holstein

[2] Corrected for the milk production during the first 6 weeks of lactation, for cows that had a second calf.

[3] Calculated from the milk production per lactating day and the length of the dry period per VWP. Differences among means are based on the milk production per lactating day.

[4] Calculated from the milk production per lactation, the days in milk and days of pregnancy at dry-off per VWP, and an average pregnancy length of 280 days, for cows pregnant at first insemination. Differences among means are based on the milk production per lactating day.

[5] Energy-corrected milk. In each VWP group, cows were milked 2 or 3 times per day.

[6] Calculated from the milk production per lactation, the average days open per VWP, and an average pregnancy length of 280 days.

Overall, primiparous cows have a lower peak and more persistent lactations compared with multiparous cows (Arbel et al., 2001; Lehmann et al., 2016; Niozas et al., 2019a). Especially in the relation between lactation length and milk production, the lactation persistency plays an important role as it affects the milk production during the prolonged period in late lactation. The greater persistency of primiparous cows compared with multiparous cows may be explained by 3 factors. First, the greater persistency may be attributed to the lower peak production of primiparous cows, which usually has a negative association with persistency (Dekkers et al., 1998). Second, the greater persistency of primiparous cows may be explained by the fact that they are still growing (Figure 2). It seems that, especially in the first 30 weeks of the lactation, the body weight of primiparous cows increased more rapidly compared with the body weight of multiparous cows. In our experiment, primiparous cows had a greater plasma insulin-like growth factor 1 (IGF-1) concentration compared with multiparous cows in the first 305 days of the lactation (Chapter 4), which may indicate more partitioning of energy toward body weight in this period of the lactation. Third, the lactation persistency is positively related with the number of mammary cells and the secretory activity (Capuco and Ellis, 2013). Possibly, this mammary survival and proliferation are affected by circulating IGF-1 (Flint and Knight, 1997). Primiparous cows had a greater serum IGF-1 concentration during early, peak, and late lactation compared with multiparous cows (Miller et al., 2006). This increased IGF-1 concentration might be related to a growing mammary gland (Weber et al., 2000). As such, the greater IGF-1 concentration for primiparous cows may contribute to their increased lactation persistency (Miller et al., 2006).

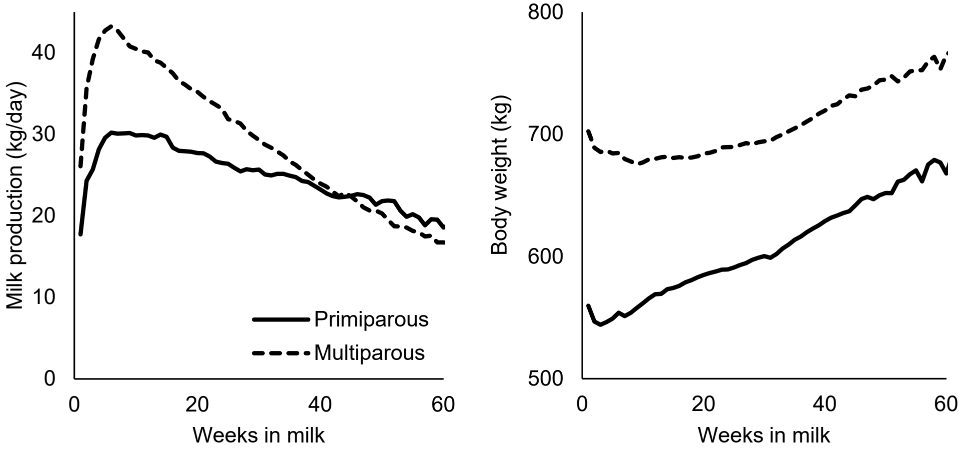


Figure 2. Milk production and body weight of primiparous and multiparous cows with a voluntary waiting period of 50 days, 125 days, or 200 days.

In addition, an extended VWP in itself could improve the lactation persistency of dairy cows. Studies that investigated the effect of pregnancy on the lactation persistency generally concluded that from 5 months onwards, pregnancy has a negative effect on the lactation persistency (Strandberg and Lundberg, 1991). This would imply that extending the VWP would result in a greater lactation persistency due to a delay in the pregnancy effect. Indeed, both in chapter 3 of this thesis and in an earlier experimental study (Niozas et al., 2019a) an extended VWP resulted in more persistent lactations for both primiparous and multiparous cows.

The lactation persistency and the effect of pregnancy on the lactation curve was also investigated on 7 farms in our network with daily milk production data between 2014 and 2018. Lactation persistency was adopted from fitted lactation curves, as the reduction in milk production per day after the peak production. A longer CInt was related with a greater lactation persistency (Figure 3). The effect of pregnancy differed per production level, where a greater production at 5 months of pregnancy was related with a stronger negative effect of pregnancy on the lactation curve at that time. As cows with a longer CInt on average had a lower milk production during that time, possibly the pregnancy effect would be less strong when pregnancy is delayed. However, the effect of pregnancy also depended on the length of the calving interval, where for cows with a longer CInt the effect of pregnancy was stronger when cows had the same production level at 5 months of pregnancy compared with cows with a shorter CInt. For primiparous cows with high production levels at 5 months of pregnancy, this effect of the length of the CInt on the pregnancy effect disappeared. Moreover, despite this effect of CInt on the pregnancy effect, the lactation persistency was increased for cows with longer CInt.

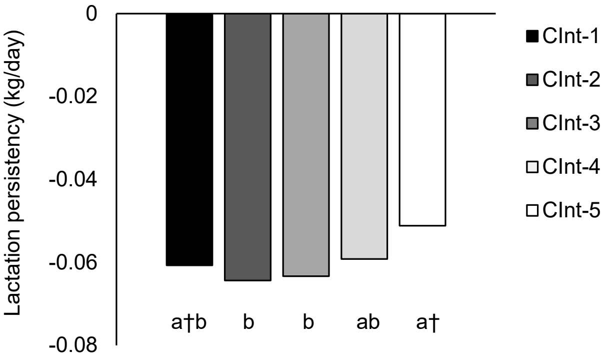


Figure 3. Lactation persistency (kg/day) of cows with different calving interval (CInt) lengths. CInt-1 <364; 364 ≤ CInt-2 < 420; 420 ≤ CInt-3 < 476; 476 ≤ CInt-4 <532; CInt-5 ≥ 532 days). Different letter means a difference among means ($P < 0.05$). Equal symbol (†) means a tendency for a difference among means ($P < 0.10$).

The 305-d milk production reflects the production potential of a cow (Kuhn and Hutchison, 2005; Kok et al., 2016). At the commercial farms in our study (Chapter 2), cows in a longer CInt had a greater 305-d production, probably mainly due to the selection by farmers of high-producing cows for longer lactations. Another explanation could be that high-producing cows had more difficulties conceiving in early lactation and therefore had an increased CInt. The cows in the longer CInt with the greatest 305-d production did not always have the greatest production per day of CInt. Primiparous cows with a CInt between 476 and 531 days had both the greatest 305-d milk production and the greatest milk production per day of CInt compared with cows with a shorter or longer CInt length. In contrast, multiparous cows with a CInt of more than 532 days had the greatest 305-d milk production, but multiparous cows with a CInt between 364 and 419 days had the greatest milk production per day of CInt. In another observational study (Lehmann et al., 2016), primiparous cows with a CInt of more than 579 days had both the greatest 305-d milk production and the greatest milk production per day of CInt compared with cows with a CInt below 519 days. In that study, although multiparous cows with a CInt of more than 579 days had the greatest 305-d milk production, the milk production per day of CInt was not different for the different CInt. This may be explained by more days in late lactation in an extended CInt, when especially multiparous cows have a lower milk production compared with early lactation.

4 Reproductive performance

When the VWP is extended, insemination is delayed to a later moment in lactation, when milk production is decreased (Gaillard et al., 2016) and cows have had more time to recover from calving and the start of the lactation. A lower milk production at the moment of insemination could be related with an improved reproductive performance (Wathes et al., 2007; Niozas et al., 2019b). In our experiment, when the VWP was extended from 50 days to 200 days, the percentage of normal ovarian cycles (duration between 18 and 24 days) around the end of the VWP was increased from 54 % to 91 %, and the number of days until pregnancy after the end of the VWP was decreased from 58 days to 31 days (Ma et al., 2020). In another recent study where the VWP was extended from 60 to 88 days, pregnancies per insemination at first service increased from 46 % to 55 % for primiparous cows, but not for multiparous cows (Stangaferro et al., 2018). Possibly, the limited extension of the VWP in that study did not give enough benefits of the lower milk production at the moment of insemination that could improve reproductive performance. When the VWP was extended from 40 to 180 days, days until pregnancy after the end of the VWP decreased from 52 to 24 days, and inseminations per pregnancy decreased from 1.77 to 1.51 (Niozas et al., 2019b). After the end of the VWP, cows had 24 (Niozas et al., 2019b) or 31 (Ma et al., 2020) days open, which may be considered as extremely few days until pregnancy. Possibly, this may be explained by the set-up of these experiments, where estrus detection systems were in use and cows were closely monitored.

The improved conception later in lactation when the VWP is extended may be explained by the negative association between metabolic status and fertility in early lactation. The decreased plasma glucose, insulin, and IGF-1 concentration and increased plasma non-esterified fatty acid (NEFA) and β-hydroxybutyrate (BHB) concentration in early lactation as a result of the NEB is associated with impaired maturation and development of oocytes (Jorritsma et al., 2004; Fouladi-Nashta et al., 2007; Leroy et al., 2008), and altered uterine environment which may impair embryonic development (Wathes et al., 2003). When cows are inseminated during or shortly after their peak production, it may be expected that milk production is still high, feed intake is limited, and cows are still in a NEB. In chapter 4, multiparous cows with a VWP of 200 days had the lowest FPCM production during the 8 weeks around the end of the VWP (30.1 kg/day) compared with cows in the shorter VWP, and FPCM production of multiparous cows with a VWP of 125 days (36.9 kg/day) was lower compared with multiparous cows with a VWP of 50 days (42.4 kg/day) during the 8 weeks around the end of the VWP. Moreover, multiparous cows with a VWP of 200 days had a greater plasma insulin and IGF-1 concentration around the end of the VWP compared with multiparous cows with a VWP of 125 or 50 days. Cows that were pregnant before 85 days in milk (DIM) had a greater plasma IGF-1 concentration during the first 7 weeks of lactation compared with cows that failed to conceive before 230 DIM (Pushpakumara et al., 2003). As such, a greater concentration of IGF-1 at the time of insemination in our experiment may have been related with the fewer days until pregnancy after an extended VWP of 200 days.

In observational studies, opposing results were reported. When investigating 51,791 first inseminations, it was reported that conception rate at first insemination was lower when insemination took place before 60 DIM compared with after 60 DIM (Inchaisri et al., 2010). In chapter 2, a longer CFSI was not associated with the number of services to conception, but cows with a CFSI between 140 and 195 days had a lower conception rate at first service compared with cows with a CFSI between 84 and 139 days. The selection of mainly high-producing cows for an extended CFSI in chapter 2 may have resulted in an equal milk production at the time of first insemination. In turn, this may have resulted in the equal reproductive performance during this time (Butler et al., 1981). Moreover, a longer CInt was associated with more services to conception and a lower conception rate at first service (Chapter 2). Possibly, the longer CInt was the result of impaired reproductive performance. In an earlier observational study, the length of the CInt was not related with conception rate (Lehmann et al., 2016).

5 Cow health

Most diseases of dairy cows are related with the start of the lactation after calving (Ingvartsen et al., 2003; Collard et al., 2000; Koeck et al., 2012). When the VWP is extended, the frequency of calving moments is reduced. This might reduce the yearly incidence of diseases of dairy cows (Knight, 2005; Lehmann et al., 2014). In addition, an extended lactation may result in a reduced milk production at the end of the lactation. Dry-off at a lower milk production might improve udder health (Rajala-Schultz et al., 2005), but more days in late lactation could be related with an increase in SCC (Miller et al., 1983). Moreover, a lower milk yield in late lactation might be related with fattening, which could increase the severity of the NEB during the start of the subsequent lactation (Schuh et al., 2019), possibly related with an increased risk for diseases.

5.1 Disease cases: per lactation and per year

The effect of an extended VWP on diseases depends on several factors. One factor is whether indeed most diseases cases occur after calving, and if the number of disease cases during the lactation would stay the same when the lactation is extended. In our experiment, in the first 6 weeks after the subsequent calving, the total number of disease cases was 2.8 per day for all cows (0.22 per cow per day), compared with 0.9 disease cases per day from 6 weeks after calving until the subsequent calving for all cows (0.006 per cow per day) (adjusted from chapter 5). This indicates that indeed the start of the lactation is a period with an increased risk for diseases compared with the complete lactation.

To our knowledge, until now occurrence of all health issues has not been reported for a complete lactation in an experiment with cows with an extended VWP. In our experimental study, disease cases were recorded for the complete lactation and the first 6 weeks of the subsequent lactation (Chapter 5). To include the calving period which may have been affected by the VWP, we included the period from 6 weeks after calving until 6 weeks after the next calving or until culling for the analysis of the effect of the VWP on disease cases. In this complete period, cows with a longer VWP had numerically more disease cases (Table 2A). This can be explained by more days in lactation for cows with an extended VWP. When expressing the disease cases per year weighted for the lactation length of a cow, cows with a shorter VWP had numerically more disease cases (Table 2B). This could indicate that indeed the diseases around calving have a larger contribution to the disease cases, and as the calving period has a relatively greater contribution to the lactation with a shorter VWP, the disease cases were increased in lactations with a shorter VWP.

For an analysis of diseases, one dataset of 153 cows on one experimental farm is too limited to draw conclusions from. Moreover, the cows in this dataset were all kept under similar management practices. It is important to note that, also in a short CInt or with a short VWP, cows can have good health with the right transition management. Environmental stressors such as changes in group composition or changes in feed composition related with the transitions around calving also affect health. As such, management related with these changes can affect the disease cases and overall health status of dairy cows (Mulligan and Doherty, 2008; Roche et al., 2013). When, however, there are indeed more disease cases in the period around calving, an extended lactation will usually result in fewer disease cases per year.

Table 2. Number of cases of a disease in the complete experimental period from wk 6 after calving until wk 6 after the next calving (A) and number of cases of a disease expressed per year and weighted for the lactation length of a cow in the experiment (B) for dairy cows with a voluntary waiting period of 50, 125, or 200 days (VWP50, VWP125, or VWP200) (adjusted from chapter 5)

A.	VWP50	VWP125	VWP200	Total
N cows	53	49	51	153
Milk fever	10	17	9	36
Ketosis	1	1	3	5
Clinical mastitis	28	25	31	84
Retained placenta	4	1	4	9
Chronic endometritis	8	8	7	23
Endometritis	1	1	2	4
Pyometra	4	3	4	11
Cystic ovaries	11	8	19	38
Abortion	0	1	1	2
Leg and claw disorders[1]	47	67	48	162
Intestine and stomach disorders	14	22	15	51
Other	8	13	13	34
Total	136	167	156	459
Disease cases per cow per lactation[2]	3.1	3.9	3.4	*P* = 0.33

B.	VWP50	VWP125	VWP200	Total
N cows	53	49	51	153
Milk fever	10	14	7	31
Ketosis	1	1	2	4
Clinical mastitis	28	21	24	73
Retained placenta	4	1	3	8
Chronic endometritis	8	7	5	20
Endometritis	1	1	2	4
Pyometra	4	3	3	10
Cystic ovaries	11	7	15	33
Abortion	0	1	1	2
Leg and claw disorders[1]	47	56	37	140
Intestine and stomach disorders	14	18	12	44
Other	8	11	10	29
Total	136	141	121	398
Disease cases per cow per year	2.6	2.8	2.4	
Disease cases per cow per year[2,3]	3.1	3.0	2.8	*P* = 0.90

[1] Including all claw disorders reported during regular checks (i.e., at 2 weeks before dry-off and between 100 and 150 days in milk).

[2] Least squares mean number of cases of all diseases per cow.

[3] Unweighted for the lactation length of a cow in the experiment.

5.2 Relation between milk production at the end of the lactation and cow health

With a short calving interval and the current high milk production, milk production at dry-off may be high, with negative consequences for the dry-off process (Rajala-Schultz et al., 2005; Odensten et al., 2007) and cow welfare (Bertulat et al., 2013; Zobel et al., 2013). Cows with a milk production of more than 18 kg/day before dry-off had higher plasma cortisol concentrations during the dry period, which may indicate stress (Odensten et al., 2007). Moreover, these cows had an increased risk of developing intra-mammary infections during the dry period, after calving, or both, compared with cows with a lower milk production at dry-off (Odensten et al., 2007). Moreover, every 5 kg higher milk production above 12.5 kg/day at dry-off increased the chance of having an intramammary infection at calving by 77 % (Rajala-Schultz et al., 2005). This increased risk for infections when milk yield is high at dry-off might be the result of the leakage of milk, which slows the formation of the protective keratin plug and allows an entry to the udder by environmental pathogens (Rajala-Schultz et al., 2005). Sudden dry-off is not a big problem for low-yielding dairy cows, but high-yielding cows experience high udder pressure and an increase in glucocorticoid production, an indication for stress, when dried-off suddenly (Bertulat et al., 2013). Milk cessation methods, such as reducing the milking frequency and adjusting the ration in the final days before dry-off, can be used to mitigate the dry-off process (Valizaheh et al., 2008; Zobel et al., 2013; Vilar and Rajala-Schultz, 2020). However, even with these strategies it may be challenging to reduce the milk production of dairy cows that still have high productions within these final days before dry-off. In chapter 3, cows with a VWP of 125 days had a lower milk production in the final 6 weeks before dry-off, and cows with a VWP of 200 days tended to have a lower milk production during that period, compared with cows with a VWP of 50 days. Moreover, cows with a VWP of 180 days had a lower milk production in the week of dry-off, and more cows in that group were dried-off below 15 kg/d, compared with cows with a VWP of 40 or 120 days (Niozas et al., 2019a). As such, an extended VWP associated with a lower milk production at the moment of dry-off might be beneficial to udder health.

However, cows that spend more days in late lactation due to an extended VWP may have an increased SCC during that time, as a reduced milk production has been related to a greater SCC (Miller et al., 1983). Nevertheless, when the VWP was extended from 40 to 120 or 180 days, no effect from VWP on SCC or mastitis cases was reported (Niozas et al., 2019a). In other studies, where cows were randomly assigned to a CInt of either 12 or 18 months, the CInt did not affect the SCC (Österman et al., 2005; Sorensen et al., 2008). Also in our experiment, the VWP did not affect the SCC or the number of clinical mastitis cases, per lactation or per year (Ma et al., in press). In these experiments, cow numbers were limited for an analysis of diseases. Moreover, only healthy cows were included based on having no severe dystocia, grade 3 metritis, or septicemic mastitis before insemination (Niozas et al., 2019a), or having no clinical mastitis or a SCC >250,000 before the previous dry-off (Ma et al., in press). Possibly, this selection of healthy cows may have resulted in overall limited levels of SCC. This does show, however, that for healthy cows an extended VWP would not be an issue in terms of SCC or mastitis cases.

In addition, the reduction in milk production at the end of the extended lactation may be related to an increase in body condition during that time (Niozas et al., 2019a). In our study, multiparous cows with an extended VWP had an increased plasma insulin and IGF-1 concentration during the pregnancy period (Chapter 4). In an earlier study, cows had elevated plasma concentrations of IGF-1, leptin, and glucose, a decreased milk yield, and an increased BW from 301 to 600 DIM compared with the period from 0 to 300 DIM, indicating more partitioning of energy toward body weight later in lactation (Marett et al., 2011). Indeed, in our study, multiparous cows with an extended VWP of 200 days had a greater BCS and greater BW gain at the end of the lactation compared with cows with a VWP of 50 days. The VWP did not affect the BCS or BW gain of primiparous cows (Chapter 3 and chapter 4). In another study, an extended VWP of 180 days was related with a greater BCS at the end of the lactation compared with a VWP of 40 or 120 days, but parity did not affect this relationship (Niozas et al., 2019a).

An increased BCS at the end of the lactation could imply an increased risk for metabolic disorders in the subsequent lactation (Gillund et al., 2001). In our study, multiparous cows with a VWP of 200 days had a greater BCS in the start of the subsequent lactation, and a more severe NEB in the first week after the subsequent calving (Chapter 4). For primiparous cows, the VWP did not affect the body condition or metabolic status at the end of the lactation or at the start of the subsequent lactation. For multiparous cows, the more severe NEB was also reflected in a greater NEFA concentration in the first 6 weeks after calving. Numerically, cows with a VWP of 200 days also had more disease cases in the first 6 weeks after the subsequent calving compared with cows with a VWP of 50 or 125 days (Table 3).

Table 3. Number of cases of a disease in the first 6 weeks after calving of dairy cows with an extended voluntary waiting period of 50, 125, or 200 days in the previous lactation (VWP50, VWP125, or VWP200) (adjusted from chapter 5)

	VWP50	VWP125	VWP200	Total
N cows	47	42	38	127
Milk fever	10	13	9	32
Ketosis	1	1	2	4
Clinical mastitis	9	3	12	24
Retained placenta	4	1	3	8
Chronic endometritis	5	5	4	14
Endometritis	1	1	2	4
Pyometra	0	1	0	1
Cystic ovaries	0	0	2	2
Abortion	0	0	0	0
Leg and claw disorders[1]	2	5	2	9
Intestine and stomach disorders	1	1	1	3
Other	2	5	8	15
Total	35	36	45	116
Disease cases per cow in the first 6 weeks of lactation[2]	0.7	0.9	1.2	$P = 0.42$

[1] Including all claw disorders reported during regular checks (i.e., at 2 weeks before dry-off and between 100 and 150 days in milk).

[2] Least squares mean number of cases of all diseases per cow.

6 Economic performance

The voluntary waiting period and the resulting lactation length may affect milk and milk content production, number of calves born, feed supply, number of inseminations, number of veterinary treatments, number of cows that are culled, number of cows that do not conceive, and farm labor (Inchaisri et al., 2011; Lehmann et al., 2016; Liang et al., 2017). These factors can all be expected to be related with the economic performance of cows. Earlier studies that modeled the consequences of an extended lactation for economic result reported reduced revenues in extended lactations, mainly due to a reduced yearly milk production (Figure 4). The reduced milk production was attributed to fewer days in peak production and more days in late lactation when milk production is lower in extended lactations.

Most modeling studies are relatively old and were caried out before 2010 or even before 2000 (Holmann et al., 1984; Schmidt, 1989; Strandberg and Oltenacu, 1989; Groenendaal et al., 2004; Inchaisri et al., 2011; Steeneveld and Hogeveen, 2012). In 1980, the average Dutch dairy cow produced 5,466 kg of milk in a lactation of on average 311 days (CRV, 2020). In 2010, this production was 9,400 kg milk in a lactation of on average 353 days. In 2020, a dairy cow produced 10,290 kg in a lactation of on average 356 days. In 2015, the milk quota were withdrawn in Europe. This means that milk production is not limited to a maximum level anymore, and a milk production above the milk quota has become profitable. Therefore, a lower milk production per year, possibly due to an extended lactation, may be expected to be more expensive compared with before 2015. However, the still increasing production level of dairy cows may affect the result of an extended VWP for milk production, as high-producing or persistent cows can maintain milk production for a longer period within one lactation, resulting in limited or no milk production losses (Kok et al., 2019).

In order to have a good economic performance in an extended lactation, maintaining milk production is important as it affects the difference between peak production and milk production at the end of the lactation. With a greater persistency, it was longer profitable to delay insemination (Dijkhuizen et al., 1985), and cows had a higher probability that an extended VWP was economically optimal (Inchaisri et al., 2011). Moreover, when persistency increased by 0.02 kg per day, multiparous cows with an extended lactation had a more limited reduction in milk production per day of CInt, and primiparous cows with an extended lactation had an increase in milk production per day of CInt (Kok et al., 2019). For primiparous cows, this increase in persistency resulted in a similar net partial cash flow (NPCF) when the lactation was extended with 4 months (Kok et al., 2019). The increase in milk production and lactation length of dairy cows in the last decennia may indicate an improved lactation persistency. Therefore, the reduced economic performance in extended lactations may be more limited than earlier estimated.

In addition, modeling studies used farm data or separate effect studies as input. This data did usually not include lactations of cows where the VWP was deliberately extended as a strategy of the farmer. Cows that are deliberately managed for an extended lactation may have production characteristics that are more suitable for an extended lactation. For example, cows with a CInt of more than 19 months had a similar milk production per day of CInt as cows with a CInt of 13 months (Lehmann et al., 2016). As a result, these cows that were managed for an extended lactation did not have a reduction in yearly NPCF when the lactation was modeled to extend with 2 or 4 months (Kok et al., 2019). Moreover, culling in observational studies is often affected by many other factors than health issues, such as the number of available replacement heifers.

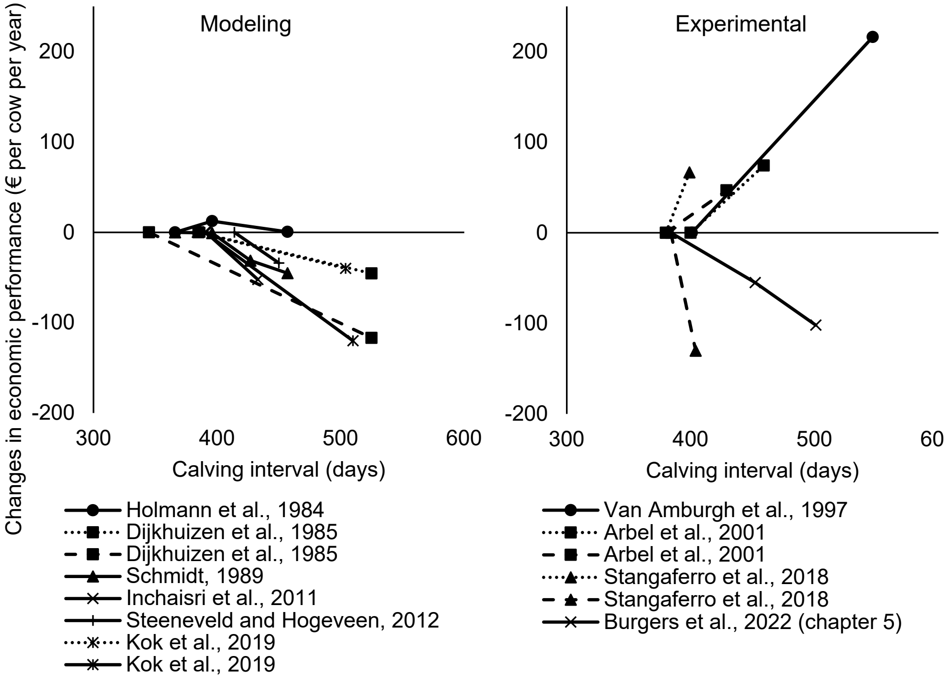


Figure 4. Changes in economic performance relative to the shortest calving interval per study, for modeling studies and experimental studies. For primiparous cows, lines are dotted. For multiparous cows, lines are striped. For all cows, lines are uninterrupted. Studies presented results in US dollars, euros, or Dutch guilders; dollars and guilders are converted to euros (€) by using the exchange rate during the publication year of the respective studies.

In correctly executed experimental studies, cows are randomly assigned to a VWP, data can be measured on individual cows, and management can be expected to have a limited effect on the differences between VWP groups. In this thesis, all input for the calculation of the NPCF was based on measured variables on cows that were blocked for parity, expected milk production, and the breeding value for persistency, and within blocks randomly assigned to a VWP of 50, 125, or 200 days (Chapter 5). As such, daily milk production of cows with a deliberately extended lactation can be compared with the milk production of cows without an extended lactation. In addition, in order to study the economic consequences, not only the milk production but also the fat, protein, and lactose production are important (Wilmink, 1988). Therefore, in the economic analysis, prices for milk, fat, protein, and lactose production were included. The total yearly revenues, based on revenues from milk, fat, protein, and lactose, and revenues from calves, were greater for cows with a VWP of 50 days compared with cows with a VWP of 200 days, mainly due to greater yearly revenues for milk and milk content production. This result was similar to earlier modeling studies, where economic performance reduced when yearly milk production was reduced in longer CInt. In our study, the lower total yearly revenues of cows with a VWP of 200 days were partially compensated by lower total yearly costs. Cows with a VWP of 200 days had lower costs for concentrates and inseminations. Moreover, costs for veterinary treatments and culling were lower for cows with a longer VWP, although the differences among the VWP groups for these costs were not significant. Still, these lower costs contributed to the total costs of the respective VWP groups. The disadvantage of an experimental study is the relatively low number of cows, which makes it difficult to analyze differences in veterinary treatments and culling.

Other experimental studies reported ambiguous results for economic performance, as illustrated in figure 4. In our study, numerically, cows with a VWP of 200 days had a €102 lower yearly NPCF compared with cows with a VWP of 50 days, and cows with a VWP of 125 days had a €47 lower yearly NPCF compared with cows with a VWP of 50 days (Chapter 5). This difference was not significant, which may be explained by the relatively low number of cows for an economic analysis. More importantly, it shows that the variation among individual cows was larger than the effect of the VWP. To investigate this variation, we divided the cows over 3 economic classes based on their yearly NPCF. Cows with a yearly NPCF of > €1,400/year had a 5 kg/day of CInt greater milk production, a 0.2 kg/day of CInt greater protein production, a 0.3 kg/day of CInt greater fat production, and a 0.2 kg/day of CInt greater lactose production compared with cows with a yearly NPCF of < €1,100/year (Chapter 5). Moreover, cows with the greater NPCF had on average 5 fewer veterinary treatments during the experiment. The VWP, CFSI, or CInt were not related with the NPCF. The effect of the economic class on lactation persistency depended on the VWP: only for cows in VWP125, the lactation persistency was greater for cows with a yearly NPCF between €1,100 and €1,400/year compared with cows with a yearly NPCF of > €1,400/year. This was in contrast with earlier modeling studies, where NPCF in extended lactations increased with an increase in persistency (Inchaisri et al., 2011; Kok et al., 2019). Possibly, in our study, the increased persistency was related to a decreased peak production (Dekkers et al., 1998). The breeding value for persistency, however, was positively related with the NPCF in our study.

Other studies also reported differences in yearly NPCF due to differences among individual cows. For example, the yearly NPCF increased numerically when the VWP was extended for primiparous cows but decreased numerically when the VWP was extended for multiparous cows (Stangaferro et al., 2018). Moreover, the yearly NPCF increased when the VWP was extended for high-producing cows (defined as a primiparous cow with once a milk production of at least 30 kg per day in the first 3 monthly records or a multiparous cow with an above herd average 305-d production in the previous lactation) (Arbel et al., 2001) or for cows whose milk production was stimulated with the use of bST (Van Amburgh et al., 1997). These studies indicate that cow characteristics such as parity and milk production affect the economic performance of individual cows. In the next part, I will discuss the implications of the large variation among individual cows.

7 Variation among cows in their response to an extended lactation

In chapter 2, data from farmers that already extend the VWP for (part of) their cows was analyzed. In that chapter, strategies to extend the VWP varied among farmers. Some farmers used a fixed VWP for the entire herd, and they did not take into account individual variation among cows. Other farmers selected individual cows for a specific VWP length, based on parity, milk yield or body condition (Chapter 2). Still, these farmers indicated that, while they did look at individual cows to decide on the VWP, they still had some difficulties with selecting the appropriate cows for an extension of the VWP.

In this thesis, individual variation among cows in terms of milk production, lactation persistency, and body condition was large, especially for multiparous cows (Figure 5). For primiparous cows, variation between individuals was relatively small. Together with their great lactation persistency and relatively equal milk production in extended lactations compared with shorter lactations, this makes primiparous cows very suitable for an extension of the VWP. However, when primiparous cows get an extended lactation, it takes more time before they become a, usually more productive, multiparous cow. Although multiparous cows might be more productive in general compared with primiparous cows (Lee and Kim, 2006), there is an increased risk for a lower production or cashflow for multiparous cows as the individual variation is larger, regardless of the lactation length. Therefore, delaying that risk may be an advantage. Multiparous cows had an increased risk for body condition loss during early lactation, disorders in early lactation, and culling due to reproductive failure compared with primiparous cows (Lee and Kim, 2006). In that respect, an extended lactation may be also beneficial for multiparous cows, to reduce these moments with high risks for diseases. For multiparous cows, lactation length can then be customized to limit risks for a reduction in cashflow or milk production or an increased body condition at the end of the lactation.

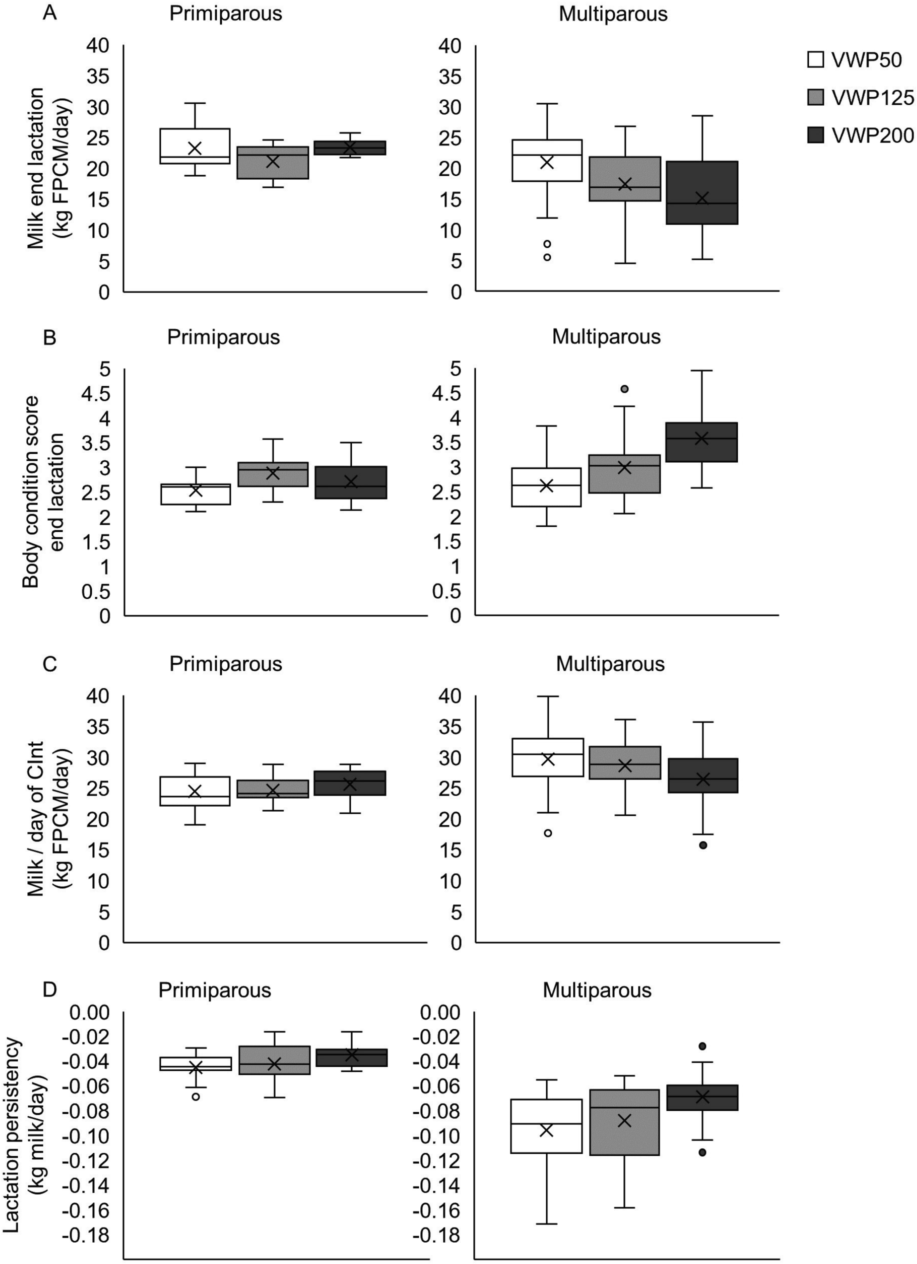


Figure 5. Fat- and protein-corrected milk production (FPCM) in the final 6 weeks of lactation (A), body condition score in the final 12 weeks of lactation (B), FPCM per day of calving interval (CInt) (C), and lactation persistency between day 100 and the start of dry-off (D) of primiparous and multiparous cows with a voluntary waiting period for insemination of 50, 125, or 200 days (VWP50, VWP125, or VWP200) that had a successful second calving in the experiment and a dry period (n=124).

Individual cow characteristics affected the economic performance of cows (Chapter 5). The cashflow is mainly affected by the milk production per day of CInt. Milk production before insemination and peak production were positively associated with milk production per day of CInt (Chapter 4). Therefore, these cows may be more suitable for an extended lactation to limit the risk for a lower cashflow. Moreover, in another study, 305-d milk production could be predicted by milk production data in the first 150 days in lactation, and cows with a 305-d milk production of on average more than 20 kg per day were considered suitable for a longer lactation (Manca et al., 2020). In an observational study where cows were selected by farmers for an extended lactation, milk production in the previous lactation and in the beginning of the current lactation were related with milk production of cows during their (extended) lactation (Lehmann et al., 2017).

Next to milk production per day of CInt, also milk production and fattening at the end of the lactation can be considered a point of attention in extended lactations. In chapter 4, multiparous cows with an extended VWP had a greater risk for an increased body condition at the end of the lactation, and a more severe NEB in the beginning of the subsequent lactations. An earlier study reported that high-producing cows had a lower risk for an increased BCS at the end of their lactation, associated with their greater milk production during the lactation (Lehmann et al., 2017). As such, high-yielding cows might have a lower risk for an increased body condition at the end of an extended lactation, and with that a lower risk for a severe NEB after the subsequent calving. In our prediction models of chapter 4, a higher milk production and a lower body condition before insemination were associated with a higher milk production and a lower body condition score at the end of the lactation. As earlier, similar relationships were reported in the prediction of different lactation performance variables (Lehmann et al., 2017), the prediction models in chapter 4 may provide a direction for selecting individual cows for an extended VWP.

8 Extended lactations in practice

The farmers in our network that already apply extended lactations on their farm were diverse, with in 2018 between 50 and 260 cows, an average 305-d FPCM production between 8,221 and 10,786 kg, and an average calving interval between 400 and 485 days (Chapter 2). As such, there seems to be not one specific type of farmer for whom this management strategy fits. However, extended lactations as a management strategy may not fit all farmers. For example, farmers may differ in fertility management, milk production expectations, and attention for individual cows. Differences in goals and intentions may affect behaviour of dairy farmers (Bergevoet et al., 2004). Moreover, for pastoral systems with a seasonal calving system, an extended lactation may imply either a one-year CInt or a two-year CInt. Some studies on pastoral systems, however, investigated the possibilities of 18-month CInt with calvings in spring or autumn (Auldist et al., 2007). When a farmer is motivated, extended lactations may suit many types of farms and systems.

8.1 Motivation of the farmer for the application of extended lactations

The motivations of the farmers in our network to extend the VWP for (part of) their herd were questioned in a survey. The motivations that the 13 farmers mentioned can be summarized in 5 aspects: 1. Improve cow health and fertility (13 farmers), 2. Benefit from high-producing cows and increased milk solids level (10 farmers), 3. Reduce (unpredictable) labor (10 farmers), 4. Reduce costs (3 farmers), and 5. Improve the sector image (2 farmers) (Van Dooren, 2019). Motivations were often not economically driven but related with job satisfaction.

One important motivation that was mentioned was the reduction of unpredictable labor. This motivation is not studied often or included in economic analyses, but 10 farmers viewed this as highly important. A calving cow is usually associated with extra labor (Boulton et al., 2017), and the moment of calving can be unpredictable. Farmers have indicated that this unpredictable labor is undesirable, as it can take a lot of time and a cow that is about to calve needs to be checked regularly and outside of usual working hours. Moreover, when calving is difficult, labor around a calving cow takes more time. In addition, this could be associated with an increased risk for diseases or death of the cow or calf, reducing the work pleasure. With an extended lactation, fewer calves are being born on the farm, so farmers experience less of these unpredictable or difficult calving moments. Moreover, fewer calves on the farm would reduce labor for calf rearing. This is especially important when calves may have to stay on the dairy farm for a longer period in the future (LNV, 2021).

8.2 Making extended lactations applicable to a larger group of cows

As discussed in the previous part, farmers can select specific cows for an extended lactation with individual cow characteristics. However, selection of specific cows can be difficult, as many factors affect the lactation performance. Therefore, when farmers want to extend the lactations of their cows, they could use specific management strategies to maintain milk production and lactation persistency of cows with an extended lactation. This could make an extended lactation applicable to a larger group of cows.

First, an increased milking frequency may stimulate a greater lactation persistency (Österman et al., 2005; Sorensen et al., 2008). Cows with a CInt of 18 months had a greater milk production per day of CInt when they were milked 3 times per day compared to when they were milked 2 times per day (Österman and Bertilsson, 2003). Moreover, numerically, cows with a CInt of 18 months had a lower milk production per day of CInt with 2 times per day milking but a greater milk production per day of CInt with 3 times per day milking compared with cows with a CInt of 12 months (Österman and Bertilsson, 2003). Next to milking 3 times per day for the complete lactation, it can be hypothesized that changing the milking frequency during the course of the lactation may be beneficial for the persistency. For example, when cows were milked once per day during the first 4 weeks of lactation, their production in the first 10 weeks of lactation was reduced compared with cows that were milked twice per day during the complete lactation (McNamara et al., 2008). As such, a lower milking frequency in early lactation may improve lactation persistency due to the negative relationship between persistency and peak production (Dekkers et al., 1998). Milking robots may enable a change in milking frequency throughout the lactation to improve lactation persistency in an extended lactation.

Second, altering dietary energy content may stimulate milk production or limit fattening. Milk production was increased more when energy supply increased with high levels of protein compared with low levels of protein, and an increased level of metabolizable protein increased the partitioning of energy toward milk production (Brun-Lafleur et al., 2010; Daniel et al., 2016). In another study, cows that were fed a full total mixed ration (TMR) diet had a greater milk production during a 670-day lactation compared with cows that grazed on pasture and were supplemented with grain and forage (13,231 vs 11,263 kg), but fewer cows with the TMR diet were able to maintain milk production until 600 DIM (Grainger et al., 2009). This indicates that a full TMR diet improved total milk production, but this milk production level was not sustained for the extended lactation. As a result, these cows gained more body condition during the lactation. In addition, a more lipogenic diet might improve lactation persistency, as it does not provide glucose precursors (Van Knegsel et al., 2007). This may reduce plasma insulin and IGF-1 concentration. A lower plasma insulin and IGF-1 concentration is associated with more energy partitioning to milk production (Hart, 1983). Indeed, a lipogenic ration was associated with more energy partitioned to milk, a lower plasma glucose concentration, and a lower BCS compared with a glucogenic ration during mid and late lactation (Mahjoubi et al., 2009). These dietary strategies could stimulate milk production and limit an increase in BCS at the end of an extended lactation.

Third, admission of bovine somatotropin (bST) could stimulate milk production in an extended lactation (Van Amburgh et al., 1997). In the EU, however, it is prohibited to treat cows with bST, as it is only used to enhance milk production, and may have adverse effects on cow health (European Commission, Council Decision 1999/879/EC of 17 December 1999). Besides these strategies, it could be hypothesized that breeding approaches could make cows more suitable for an extended lactation. For example, in the prediction models of chapter 4, the breeding value for persistency was positively related with the milk production per day of CInt and the milk production in the final 6 weeks of lactation. In addition, some farmers in our network used the breeding value for milk production and protein content to breed suitable cows for an extended lactation. As such, appropriate breeding decisions could make extended lactations applicable for more cows.

8.3 Consequences for calves

When insemination of cows is delayed, metabolism at the time of conception and during pregnancy was different (Chapter 4). Parental factors around conception and during pregnancy, such as body composition and metabolism, may affect the development and health status of the offspring, both in early life and later in life (Fleming et al., 2018). Therefore, it may be expected that the difference in metabolism during conception and pregnancy of cows with different VWP can affect the health and performance of the calf. Until now, however, the relation between cows with extended lactations and the health and performance of their offspring has not been studied. Moreover, an extended lactation could affect the colostrum quality after the subsequent calving. For example, days open, lactating days, and milk production in the previous lactation were positively correlated with the immunoglobulin G (IgG) concentration in colostrum (Cabral et al., 2016). Moreover, body condition at the end of the lactation was affected by the lactation length (Chapter 3 and 4), and body condition before calving was positively correlated with the immunoglobulin concentration in colostrum (Zhao et al., 2019). In our experiment, however, the VWP in the previous lactation did not affect the IgG concentration in colostrum (VWP50: 153 mg/mL, VWP125: 125 mg/mL, VWP200: 128 mg/mL; P=0.24), the IgM concentration in colostrum (VWP50: 6.9 mg/mL, VWP125: 5.4 mg/mL, VWP200: 5.3 mg/mL; P=0.12), or the colostrum quantity (VWP50: 5.1 L, VWP125: 6.1 L, VWP200: 5.5 L; P=0.45). Therefore, it may be expected that an extension of the VWP has limited effects on colostrum quality.

When the calving interval is extended, fewer calves will be born on dairy farms. At the moment, around 30% of calves are kept on the dairy farm for replacement, and the other surplus calves usually are transferred to the veal industry (CRV, 2020; Bokma et al., 2020). The welfare and health of these surplus calves with regards to transport or care are a growing public concern (Bolton and Von Keyserlingk, 2021). Therefore, extended lactations, resulting in fewer calves, may result in a better match between youngstock and replacement, and improve the image of the dairy sector. Still, sufficient calves for replacement should be available. Regarding that, a remaining question is how to handle the fact that with selection of cows that are the most suited for an extended lactation, these cows get the fewest calves in the herd. One study reported, however, that the effect of an extended lactation on the genetic lag (i.e., the difference between genetic level of semen bulls and producing cows) is small (Clasen et al., 2019). In addition, the use of sexed semen could limit this genetic lag due to an extended lactation, (Clasen et al., 2019).

8.4 Consequences for the lifespan of dairy cows

Extended lactations can be related with the lifespan of dairy cows in 2 ways. First, an extended lactation could be directly related to inseminating cows with difficulties to conceive multiple times, resulting in fewer culling. Second, it may be expected that, with a reduction in the frequency of transitions around calving due to extended lactations, diseases related with the start of lactation are reduced. This could increase lifespan of dairy cows, as risk for culling due to diseases is greatest in the first 60 days of lactation (Pinedo et al., 2014). In a study on 2,574 herds, around 5000 cows died or were culled in the first week of lactation, against less than 3000 cows in all other weeks of the lactation (Dechow and Goodling, 2008). However, most culling due to a low milk production or issues with breeding happens after 330 days in lactation (Pinedo et al., 2014). Culling in early lactation can be considered extra undesirable, as the high costs made during the rearing period or the dry period are not regained by the expected milk production of the cow (Dechow and Goodling, 2008). Farmers mention fewer calvings and a related increase in lifespan as one of the motivations to extend the lactation. The relation between lifespan and lactation length is, however, still unclear. When investigating the lifespan of dairy cows on 11 farms that already extended the lactation for (part of) their herd, a longer CInt was associated with an increased lifespan, and a longer final lactation length. This increased lifespan may be the result of the extended lactations and consequential fewer transition periods. However, an extended CInt in retrospective studies is often confounded with either milk production or impaired health or fertility in early lactation, affecting the culling of cows. Experimental studies are often limited in terms of animal numbers and length of the study to draw conclusions on culling and lifespan. A large and long-term controlled study may be able to demonstrate the relation between VWP and lifespan of dairy cows. Such a study, however, may be not very realistic due to the required resources.

8.5 Consequences for the environmental impact

An extended lactation may affect the environmental impact, as lactation length is related with milk production, number of calves born, replacement rate, and changes in feeding (Kok et al., 2019; Lehmann et al., 2019; Sehested et al., 2019). As emissions of greenhouse gasses are usually expressed per kg of milk, emissions are increased when milk production is decreased. A rise in lactation length was shown to be related with an increase in greenhouse gas emissions (Wall et al., 2012). However, an increased milk production had a larger effect on reducing greenhouse gasses than changes to the lactation length, replacement rate, or persistency. As such, when milk production is not decreased or even increased in extended lactations due to selection of suitable cows, greenhouse gasses might not be affected or reduced.

9 Conclusions

This thesis evaluated the consequences of an extended lactation for milk production, reproductive performance, health, metabolism, and economic performance. Consequences of extended lactations were studied both on commercial dairy farms and in an experimental setting. Overall, longer lactations were related with a decrease in milk production per day of CInt, due to relatively fewer days in peak production and more days in late lactation. Moreover, longer lactations were related with a decreased milk production at the end of the lactation, an increased body condition at the end of the lactation, and a more severe NEB after the subsequent calving.

The reduced milk production resulted in lower total revenues for cows with a VWP of 200 days compared with cows with a VWP of 50 days. However, these lower total yearly revenues were partly compensated by lower total yearly costs for cows with a VWP of 200 days, mainly due to lower costs for concentrates and inseminations. As a result, the yearly net partial cashflow was not significantly related with the VWP. This may be partially attributed to the large variation among individual cows. Cow characteristics that determined the yearly net partial cashflow were milk and milk content production, and number of disease cases.

This variation among individual cows in their response to an extended lactation may enable the selection of individual cows for an extended lactation. For primiparous cows, an extended lactation did not affect the milk production or the body condition. Moreover, cows with a greater milk production before insemination, a greater peak production, and a lower body condition before insemination may be more suitable for an extended lactation, as these characteristics were related with a greater milk production at the end of the lactation, a lower body condition at the end of the lactation, and a greater milk production per day of CInt.

The main motivation of farmers that already extend the lactation for (part of) their herd was an improved cow health due to fewer calving moments. These farmers have specific strategies to select cows for an extended lactation, and this thesis can support farmers in the selection of cows. Individually customized lactation lengths may contribute to the sustainability of dairy farming, as it reduces the frequency of calving moments while limiting the risks for fattening or low milk production in extended lactations. Next to the selection of specific cows, management strategies with regard to feeding, milking, or breeding may be used to make extended lactations applicable for a larger group of cows.

References

Adamiak, S.J., K. Mackie, R.G. Watt, R. Webb, and K.D. Sinclair. 2005. Impact of nutrition on oocyte quality: cumulative effects of body composition and diet leading to hyperinsulinemia in cattle. Biol. Reprod. 73:918-926.

Allore, H.G., and H.N. Erb. 2000. Simulated effects on dairy cattle health of extending the voluntary waiting period with recombinant bovine somatotropin. Prev. Vet. Med. 46:29- 50.

Appuhamy, J.A.D.R.N., B.G. Cassell, C.D. Dechow, and J.B. Cole. 2007. Phenotypic relationships of common health disorders in dairy cows to lactation persistency estimated from daily milk weights. J. Dairy Sci. 90:4424–4434.

Arbel, R., Y. Bigun, E. Ezra, H. Sturman, and D. Hojman. 2001. The effect of extended calving intervals in high lactating cows on milk production and profitability. J. Dairy Sci. 84:600–608.

Auldist, M.J., G. O'brien, D. Cole, K.L. Macmillan, and C. Grainger. 2007. Effects of varying lactation length on milk production capacity of cows in pasture-based dairying systems. J. Dairy Sci. 90:3234-3241.

Bergevoet, R.H., C.J.M. Ondersteijn, H.W. Saatkamp, C.M.J. van Woerkum, and R.B.M. Huirne. 2004. Entrepreneurial behaviour of Dutch dairy farmers under a milk quota system: goals, objectives and attitudes. Agric. Syst. 80:1-21.

Bertilsson, J., B. Berglund, G. Ratnayake, K. Svennersten-Sjaunja, and H. Wiktorsson. 1997. Optimising lactation cycles for the high-yielding dairy cow. A European perspective. Livest. Prod. Sci. 50:5–13.

Bertulat, S., C. Fischer-Tenhagen, V. Suthar, E. Möstl, N. Isaka, and W. Heuwieser. 2013. Measurement of fecal glucocorticoid metabolites and evaluation of udder characteristics to estimate stress after sudden dry-off in dairy cows with different milk yields. J. Dairy Sci. 96:3774-3787.

Bokma, J., R. Boone, P. Deprez, and B. Pardon. 2020. Herd-level analysis of antimicrobial use and mortality in veal calves: do herds with low usage face higher mortality? J. Dairy Sci. 103:909-914.

Bolton, S.E., and M.A.G. von Keyserlingk. 2021. The Dispensable Surplus Dairy Calf: Is This Issue a "Wicked Problem" and Where Do We Go From Here? Fron. Vet. Sci. 8:347.

Bormann, J., G.R. Wiggans, T. Druet, and N. Gengler. 2002. Estimating effects of permanent environment, lactation stage, age, and pregnancy on test-day yield. J. Dairy Sci. 85:263-e1.

Boulton, A.C., J. Rushton, and D.C. Wathes. 2017. An empirical analysis of the cost of rearing dairy heifers from birth to first calving and the time taken to repay these costs. Animal 11:1372-1380.

Brotherstone, S., R. Thompson, and I.M.S. White. 2004. Effects of pregnancy on daily milk yield of Holstein–Friesian dairy cattle. Livest. Prod. Sci. 87:265–269.

Brun-Lafleur, L., L. Delaby, F. Husson, and P. Faverdin. 2010. Predicting energy × protein interaction on milk yield and milk composition in dairy cows. J. Dairy Sci. 93:4128-4143.

Burgers, E.E.A., A. Kok, R.M.A. Goselink, H. Hogeveen, B. Kemp, and A.T.M. van Knegsel. 2021a. Fertility and milk production on commercial dairy farms with customized lactation lengths. J. Dairy Sci. 104:443–458.

Burgers, E.E.A., A. Kok, R.M.A. Goselink, H. Hogeveen, B. Kemp, and A.T.M. van Knegsel. 2021b. Effect of extended voluntary waiting period from calving until first insemination on body condition, milk yield and lactation persistency. J. Dairy Sci. 104:8009–8022.

Burgers, E.E.A., A. Kok, R.M.A. Goselink, H. Hogeveen, B. Kemp, and A.T.M. van Knegsel. 2022. Revenues and costs of dairy cows with different voluntary waiting periods based on data of a randomized control trial. J. Dairy Sci. 105:4171-4188.

Butler, S.T., A.L. Marr, S.H. Pelton, R.P. Radcliff, M.C. Lucy, and W.R. Butler. 2003. Insulin restores GH responsiveness during lactation-induced negative energy balance in dairy cattle: effects on expression of IGF-I and GH receptor 1A. J. Endocrinol. 176:205–217.

Butler, S.T., L. Shalloo, and J.J. Murphy. 2010. Extended lactations in a seasonal-calving pastoral system of production to modulate the effects of reproductive failure. J. Dairy Sci. 93:1283-1295.

Butler, W.R., R.W. Everett, and C.E. Coppock. 1981. The relationships between energy balance, milk production and ovulation in postpartum Holstein cows. J. Anim. Sci. 53:742–748.

Butler, W.R. 2000. Nutritional interactions with reproductive performance in dairy cattle. Anim. Reprod. Sci. 60:449–457.

Butler, W.R. 2005. Nutrition, negative energy balance and fertility in the postpartum dairy cow. Cattle Pract. 13:13-18.

Cabral, R.G., C.E. Chapman, K.M. Aragona, E. Clark, M. Lunak, and P.S. Erickson. 2016. Predicting colostrum quality from performance in the previous lactation and environmental changes. J. Dairy Sci. 99:4048-4055.

Capuco, A.V., and S.E. Ellis. 2013. Comparative aspects of mammary gland development and homeostasis. Annu. Rev. Anim. Biosci., 1:179-202.

Carvalho, P.D., A.H. Souza, M.C. Amundson, K.S. Hackbart, M.J. Fuenzalida, M.M. Herlihy, H. Ayres, A.R. Dresch, L.M. Vieira, J.N. Guenther, R.R. Grummer, P.M. Fricke, R.D. Shaver, and M.C. Wiltbank. 2014. Relationships between fertility and post-partum changes in body condition and body weight in lactating dairy cows. J. Dairy Sci. 97:3666–3683.

Chebel, R.C., J.E. Santos, J.P. Reynolds, R.L. Cerri, S.O. Juchem, and M. Overton. 2004. Factors affecting conception rate after artificial insemination and pregnancy loss in lactating dairy cows. Anim. Reprod. Sci. 84:239–255.

Chen, J., N.M. Soede, H.A. van Dorland, G.J. Remmelink, R.M. Bruckmaier, B. Kemp, and A.T.M. van Knegsel. 2015. Relationship between metabolism and ovarian activity in dairy cows with different dry period lengths. Theriogenology 84:1387-1396.

Chen, J., A. Kok, G.J. Remmelink, J.J. Gross, R.M. Bruckmaier, B. Kemp, and A.T.M. van Knegsel. 2016. Effects of dry period length and dietary energy source on lactation curve characteristics over 2 subsequent lactations. J. Dairy Sci. 99:9287–9299.

Clasen, J.B., J.O. Lehmann, J.R. Thomasen, S. Østergaard, and M. Kargo. 2019. Combining extended lactation with sexed semen in a dairy cattle herd: Effect on genetic and total economic return. Livest. Sci. 223:176-183.

Collard, B.L., P.J. Boettcher, J.M. Dekkers, D. Petitclerc, and L.R. Schaeffer. 2000. Relationships between energy balance and health traits of dairy cattle in early lactation. J. Dairy Sci. 83:2683-2690.

CRV. 2019. International Dutch Cattle Improvement Co-operative. Jaarstatistieken 2018. https://www.cooperatie-crv.nl/downloads/cooperatie-crv/crv-jaarverslagen/. Visited 10-8-2019.

CRV. 2020. International Dutch Cattle Improvement Co-operative. https://www.cooperatie-crv.nl/wp-content/uploads/2020/04/E_19-Levensduur-April-2020.pdf. Visited 12-6-2022.

CRV. 2021. International Dutch Cattle Improvement Co-operative. Jaarstatistieken 2020. https://www.cooperatie-crv.nl/downloads/cooperatie-crv/crv-jaarverslagen/. Visited 19-5-2022.

Cutullic, E., L. Delaby, Y. Gallard, and C. Disenhaus. 2012. Towards a better understanding of the respective effects of milk yield and body condition dynamics on reproduction in Holstein dairy cows. Animal 6:476–487.

CVB. 2012. Cvb tabellenboek veevoeding (feedstuff table 2012), Centraal Veevoeder Bureau Lelystad, the Netherlands.

CVB. 2016. Cvb tabellenboek veevoeding (feedstuff table 2016), Centraal Veevoeder Bureau Lelystad, the Netherlands.

Daniel, J.B., N.C. Friggens, P. Chapoutot, H. van Laar, and D. Sauvant. 2016. Milk yield and milk composition responses to change in predicted net energy and metabolizable protein: a meta-analysis. Animal 10:1975-1985.

De Vries, M.J., S. van der Beek, L.M.T.E. Kaal-Lansbergen, W. Ouweltjes, and J.B.M. Wilmink. 1999. Modeling of energy balance in early lactation and the effect of energy deficits in early lactation on first detected estrus postpartum in dairy cows. J. Dairy Sci. 82:1927-1934.

Dechow, C.D., and R.C. Goodling. 2008. Mortality, culling by sixty days in milk, and production profiles in high-and low-survival Pennsylvania herds. J. Dairy Sci. 91:4630-4639.

Dekkers, J.C.M., J.H. Ten Hag, and A. Weersink. 1998. Economic aspects of persistency of lactation in dairy cattle. Livest. Prod. Sci. 53:237–252.

Delany, K.K., K.L. Macmillan, C. Grainger, P.C. Thomson, D. Blache, K.R. Nicholas, and M.J. Auldist. 2010. Blood plasma concentrations of metabolic hormones and glucose during extended lactation in grazing cows or cows fed a total mixed ration. J. Dairy Sci. 93:5913-5920.

Dijkhuizen, A.A., J. Stelwagen, and J.A. Renkema. 1985. Economic aspects of reproductive failure in dairy cattle. I. Financial loss at farm level. Prev. Vet. Med. 3:251-263.

Dillon, P., S. Crosse, G. Stakelum, and F. Flynn. 1995. The effect of calving date and stocking rate on the performance of spring-calving dairy cows. Grass Forage Sci. 50:286-299.

Dobson, H., R.F. Smith, M.D. Royal, C.H. Knight, and I.M. Sheldon. 2007. The high-producing dairy cow and its reproductive performance. Reprod. Domest. Anim. 42:17-23.

Erb, R.E., M.M. Goodwin, R.A. Morrison, and A.O. Shaw. 1952. Lactation studies. I. Effect of gestation. J. Dairy Sci. 35:224-233.

Esposito, G., P.C. Irons, E.C. Webb, and A. Chapwanya. 2014. Interactions between negative energy balance, metabolic diseases, uterine health and immune response in transition dairy cows. Anim. Reprod. Sci. 144:60–71.

European Commission. 1999. Council Decision of 17 December 1999 concerning the placing on the market and administration of bovine somatotropin (bST) and repealing Decision 90/218/EEC. Off. J. Eur. Commun.. L331:71–72.

Ferguson, J.D., D.T. Galligan, and N. Thomsen. 1994. Principal descriptors of body condition in Holstein cows. J. Dairy Sci. 77:2695–2703.

Fetrow, J., K.V. Nordlund, and H.D. Norman. 2006. Invited review: Culling: Nomenclature, definitions, and recommendations. J. Dairy Sci. 89:1896–1905.

Fleming, T.P., A.J. Watkins, M.A. Velazquez, J.C. Mathers, A.M. Prentice, J. Stephensen, M. Barker, R. Saffery, C.S. Yajnik, J.J. Eckert, M.A. Hanson, T. Forrester, P.D. Gluckman, and K.M. Godfrey. 2018. Origins of lifetime health around the time of conception: causes and consequences. Lancet 391:1842-1852.

Flint, D.J., and C.H. Knight. 1997. Interactions of prolactin and growth hormone (GH) in the regulation of mammary gland function and epithelial cell survival. J. Mammary Gland Biol. Neoplasia 2:41-48.

Fouladi-Nashta, A.A., C.G. Gutierrez, J.G. Gong, P.C. Garnsworthy, and R. Webb. 2007. Impact of dietary fatty acids on oocyte quality and development in lactating dairy cows. Biol. Reprod. 77:9–17.

FrieslandCampina. 2020. Average milk price 2015-2020. Accessed Jan. 15, 2021. https://www.frieslandcampina.com/our-farmers/owned-by-farmers/guaranteed-milk-price.

Friggens, N.C., G.C. Emmans, and R.F. Veerkamp. 1999. On the use of simple ratios between lactation curve coefficients to describe parity effects on milk production. Livest. Prod. Sci. 62:1–13.

Friggens, N.C., J.B. Andersen, T. Larsen, O. Aaes, and R.J. Dewhurst. 2004. Priming the dairy cow for lactation: A review of dry cow feeding strategies. Anim. Res. 53:453–473.

Friggens, N.C., C. Disenhaus, and H.V. Petit. 2010. Nutritional sub-fertility in the dairy cow: towards improved reproductive management through a better biological understanding. Animal 4:1197-1213.

Gaillard, C., H. Barbu, M.T. Sørensen, J. Sehested, H. Callesen, and M. Vestergaard. 2016. Milk yield and estrous behavior during eight consecutive estruses in Holstein cows fed standardized or high energy diets and grouped according to live weight changes in early lactation. J. Dairy Sci. 99: 3134-3143.

Garnsworthy, P.C., A.A. Fouladi-Nashta, G.E. Mann, K.D. Sinclair, and R. Webb. 2009. Effect of dietary-induced changes in plasma insulin concentrations during the early post partum period on pregnancy rate in dairy cows. Reprod. 137: 759-768.

Garverick, H.A., M.N. Harris, R. Vogel-Bluel, J.D. Sampson, J. Bader, W.R. Lamberson, J.N. Spain, M.C. Lucy, and R.S. Youngquist. 2013. Concentrations of nonesterified fatty acids and glucose in blood of periparturient dairy cows are indicative of pregnancy success at first insemination. J. Dairy Sci. 96:181–188.

Gillund, P., O. Reksen, Y.T. Gröhn, and K. Karlberg. 2001. Body condition related to ketosis and reproductive performance in Norwegian dairy cows. J. Dairy Sci. 84: 1390-1396.

Gobikrushanth, M., A. De Vries, J.E.P. Santos, C.A. Risco, and K.N. Galvão. 2014. Effect of delayed breeding during the summer on profitability of dairy cows. J. Dairy Sci. 97:4236–4246.

Goff, J.P., and R.L. Horst. 1997. Physiological changes at parturition and their relationship to metabolic disorders. J. Dairy Sci. 80:1260-1268.

Grainger, C., M.J. Auldist, G. O’Brien, K.L. Macmillan, and C. Culley. 2009. Effect of type of diet and energy intake on milk production of Holstein-Friesian cows with extended lactations. J. Dairy Sci. 92:1479-1492.

Green, M.J., A.J. Bradley, G.F. Medley, and W.J. Browne. 2008. Cow, farm, and herd management factors in the dry period associated with raised somatic cell counts in early lactation. J. Dairy Sci. 91:1403-1415.

Groenendaal, H., D.T. Galligan, and H.A. Mulder. 2004. An economic spreadsheet model to determine optimal breeding and replacement decisions for dairy cattle. J. Dairy Sci. 87:2146–2157.

Gross, J., H.A. van Dorland, F.J. Schwarz, and R.M. Bruckmaier. 2011. Endocrine changes and liver mRNA abundance of somatotropic axis and insulin system constituents during negative energy balance at different stages of lactation in dairy cows. J. Dairy Sci. 94: 3484-3494.

Hackmann, T.J., and J.N. Spain. 2010. Invited review: ruminant ecology and evolution: perspectives useful to ruminant livestock research and production. J. Dairy Sci. 93:1320-1334.

Hart, I.C. 1983. Endocrine control of nutrient partition in lactating ruminants. Proc. Nutr. Soc. 42:181-194.

Holman, A., J. Thompson, J. Routly, J. Cameron, D. Jones, D. Grove-White, R. Smith, and H. Dobson. 2011. Comparison of oestrus detection methods in dairy cattle. Vet. Rec. 169:47.

Holmann, F.J., C.R. Shumway, R.W. Blake, R.B. Schwart, and E.M. Sudweeks. 1984. Economic value of days open for Holstein cows of alternative milk yields with varying calving intervals. J. Dairy Sci. 67:636–643.

Hostens, M., J. Ehrlich, B. van Ranst, and G. Opsomer. 2012. On-farm evaluation of the effect of metabolic diseases on the shape of the lactation curve in dairy cows through the milkbot lactation model. J. Dairy Sci. 95:2988–3007.

Inchaisri, C., R. Jorritsma, P.L. Vos, G.C. van der Weijden, and H. Hogeveen. 2010. Economic consequences of reproductive performance in dairy cattle. Theriogenology 74:835-846.

Inchaisri, C., R. Jorritsma, P.L.A.M. Vos, G.C. van der Weijden, and H. Hogeveen. 2011. Analysis of the economically optimal voluntary waiting period for first insemination. J. Dairy Sci. 94:3811–3823.

Inchaisri, C., A. De Vries, R. Jorritsma, and H. Hogeveen. 2012. Improved knowledge about conception rates influences the decision to stop insemination in dairy cows. Reprod. Domest. Anim. 47:820–826.

Ingvartsen, K.L., R.J. Dewhurst, and N.C. Friggens. 2003. On the relationship between lactational performance and health: is it yield or metabolic imbalance that cause production diseases in dairy cattle? A position paper. Livest. Prod. Sci. 83:277-308.

ISO. 2013. ISO standard 9622: Milk and liquid milk products. Guidelines for the application of mid-infrared spectrometry. 2:14. International Organization of Standardization.

Jorritsma, R., M.L. César, J.T. Hermans, C.L.J.J. Kruitwagen, P.L.A.M. Vos, and T.A.M. Kruip. 2004. Effects of non-esterified fatty acids on bovine granulosa cells and developmental potential of oocytes in vitro. Anim. Reprod. Sci. 81:225–235.

Kay, J.K., C.V.C. Phyn, J.R. Roche, and E.S. Kolver. 2009. Extending lactation in pasture-based dairy cows. II: Effect of genetic strain and diet on plasma hormone and metabolite concentrations. J. Dairy Sci. 92:3704-3713.

Kimura, K.A.Y.O.K.O., T.A. Reinhardt, and J.P. Goff. 2006. Parturition and hypocalcemia blunts calcium signals in immune cells of dairy cattle. J. Dairy Sci. 89:2588-2595.

Knight, C.H. 2001. Lactation and gestation in dairy cows: flexibility avoids nutritional extremes. Proc. Nutr. Soc. 60:527-537.

Knight, C.H. 2005. Extended lactation: Turning theory into reality. Adv. Dairy Technol. 17:113–123.

Koeck, A., F. Miglior, D.F. Kelton, and F.S. Schenkel. 2012. Health recording in Canadian Holsteins: Data and genetic parameters. J. Dairy Sci. 95:4099-4108.

Kok, A., C.E. van Middelaar, B. Engel, A.T.M. van Knegsel, H. Hogeveen, B. Kemp, and I.J.M. de Boer. 2016. Effective lactation yield: A measure to compare milk yield between cows with different dry period lengths. J. Dairy Sci. 99:2956–2966.

Kok, A., J.O. Lehmann, B. Kemp, H. Hogeveen, C.E. van Middelaar, I.J.M. de Boer, and A.T.M. van Knegsel. 2019. Production, partial cash flows and greenhouse gas emissions of simulated dairy herds with extended lactations. Animal 13:1074–1083.

Kolver, E.S., J. Roche, C. Burke, J. Kay, and P. Aspin. 2007. Extending lactation in pasture-based dairy cows: I. Genotype and diet effect on milk and reproduction. J. Dairy Sci. 90:5518–5530.

Koprowski, J.A. and H. Allen Tucker. 1973. Bovine serum growth hormone, corticoids and insulin during lactation. Endocrinology 93: 645-651.

Kuhn, M.T., and J. Hutchison. 2005. Methodology for estimation of days dry effects. J. Dairy Sci. 88:1499–1508.

KWIN-V. 2020. Kwantitatieve Informatie Veehouderij 2020–2021 (Quantitative livestock farming information 2020–2021). Livestock Research, Wageningen University & Research.

Larsson, B., and B. Berglund. 2000. Reproductive performance in cows with extended calving interval. Reprod. Domest. Anim. 35:277–279.

Lean, I.J., J.C. Galland, and J.L. Scott. 1989. Relationships between fertility, peak milk yields and lactational persistency in dairy cows. Theriogenology 31:1093–1103.

Lee, J.Y., and I.H. Kim. 2006. Advancing parity is associated with high milk production at the cost of body condition and increased periparturient disorders in dairy herds. J. Vet. Sci. 7:161–166.

Lehmann, J.O., L. Mogensen, and T. Kristensen. 2014. Extended lactations may improve cow health, productivity and reduce greenhouse gas emissions from organic dairy production. Org. Agric. 4:295–299.

Lehmann, J.O., J.G. Fadel, L. Mogensen, T. Kristensen, C. Gaillard, and E. Kebreab. 2016. Effect of calving interval and parity on milk yield per feeding day in danish commercial dairy herds. J. Dairy Sci. 99:621–633.

Lehmann, J.O., L. Mogensen, and T. Kristensen. 2017. Early lactation production, health, and welfare characteristics of cows selected for extended lactation. J. Dairy Sci. 100:1487–1501.

Lehmann, J.O., L. Mogensen, and T. Kristensen. 2019. Extended lactations in dairy production: Economic, productivity and climatic impact at herd, farm and sector level. Livest. Sci. 220:100-110.

Leroy, J.L.M.R., T. Vanholder, G. Opsomer, A. van Soom, and A. de Kruif. 2006. The in vitro development of bovine oocytes after maturation in glucose and β-hydroxybutyrate concentrations associated with negative energy balance in dairy cows. Reprod. Domest. Anim. 41:119–123.

Leroy, J.L.M.R., A. van Soom, G. Opsomer, I.G.F. Goovaerts, and P.E.J. Bols. 2008. Reduced fertility in high-yielding dairy cows: are the oocyte and embryo in danger? Part II mechanisms linking nutrition and reduced oocyte and embryo quality in high-yielding dairy cows. Reprod. Domest. Anim. 43:623-632.

Liang, D., L.M. Arnold, C.J. Stowe, R.J. Harmon, and J.M. Bewley. 2017. Estimating US dairy clinical disease costs with a stochastic simulation model. J. Dairy Sci. 100:1472-1486.

LNV. 2021. Ministerie van Landbouw, Natuur, en Voedselkwaliteit: Scenariostudie Kalverketen - scenario's voor een andere inrichting van de keten. https://open.overheid.nl/repository/ronl-8fd16148-f322-46ab-aeb7-2ca96da03b4a/1/pdf/scenariostudie-kalverketen.pdf. Visited 24-5-2022.

Lopez, H., L.D. Satter, and M.C. Wiltbank. 2004. Relationship between level of milk production and estrous behavior of lactating dairy cows. Anim. Reprod. Sci. 81:209–223.

Lucy, M.C. 2001. Reproductive loss in high-producing dairy cattle: where will it end? J. Dairy Sci. 84:1277-1293.

Ma, J., E.E.A. Burgers, T.J.G.M. Lam, B. Kemp, and A.T.M. van Knegsel. 2020. Consequences of extending the voluntary waiting period on ovarian cyclicity in dairy cows. Page 323 in Book of Abstracts of the 71st Annual Meeting of the EAAP, session 27, poster 14.

Ma, J., A. Kok, R.M.A. Goselink, T.J.G.M. Lam, B. Kemp, and A.T.M. van Knegsel. In press. Udder health of dairy cows with an extended voluntary waiting period from calving until the first insemination. J. Dairy Res.

Mahjoubi, E., H. Amanlou, D. Zahmatkesh, M.G. Khan, and N. Aghaziarati. 2009. Use of beet pulp as a replacement for barley grain to manage body condition score in over-conditioned late lactation cows. Anim. Feed Sci. Technol. 153:60-67.

Manca, E., A. Cesarani, N.P. Macciotta, A.S. Atzori, G. Pulina, and C. Dimauro. 2020. Use of discriminant statistical procedures for an early detection of persistent lactations in dairy cows. Comput. Electron. Agric. 176:105657.

Marett, L.C., M.J. Auldist, C. Grainger, W.J. Wales, D. Blache, K.L. Macmillan, and B.J. Leury. 2011. Temporal changes in plasma concentrations of hormones and metabolites in pasture-fed dairy cows during extended lactation. J. Dairy Sci. 94:5017-5026.

Marett, L.C., M.J. Auldist, W.J. Wales, K.L. Macmillan, F.R. Dunshea, and B.J. Leury. 2019. Responses to metabolic challenges in dairy cows with high or low milk yield during an extended lactation. J. Dairy Sci. 102:4590-4605.

McAtee, J.W. and A. Trenkle. 1971. Metabolic regulation of plasma insulin levels in cattle. J. Anim. Sci. 33:438-442.

McNamara, S., J.J. Murphy, F.P. O'mara, M. Rath, and J.F. Mee. 2008. Effect of milking frequency in early lactation on energy metabolism, milk production and reproductive performance of dairy cows. Livest. Sci. 117:70-78.

Mellado, M., J.M. Flores, A. de Santiago, F.G. Veliz, U. Macías-Cruz, L. Avendaño-Reyes, and J.E. García. 2016. Extended lactation in high-yielding Holstein cows: Characterization of milk yield and risk factors for lactations > 450 days. Livest. Sci. 189:50–55.

Miller, R.H., U. Emanuelsson, E. Persson, L. Brolund, J. Philipsson, and H. Funke. 1983. Relationships of milk somatic cell counts to daily milk yield and composition. Acta Agric. Scand. A. Anim. Sci. 33:209-223.

Miller, N., L. Delbecchi, D. Petitclerc, G.F. Wagner, B.G. Talbot, and P. Lacasse. 2006. Effect of stage of lactation and parity on mammary gland cell renewal. J. Dairy Sci. 89:4669-4677.

Mohd Nor, N., W. Steeneveld, M.C.M. Mourits, and H. Hogeveen. 2012. Estimating the costs of rearing young dairy cattle in the Netherlands using a simulation model that accounts for uncertainty related to diseases. Prev. Vet. Med. 106:214–224.

Mohd Nor, N., W. Steeneveld, and H. Hogeveen. 2014. The average culling rate of Dutch dairy herds over the years 2007 to 2010 and its association with herd reproduction, performance and health. J. Dairy Res. 81:1–8.

Morrow, D.A. 1976. Fat cow syndrome. J. Dairy Sci. 59:1625-1629.

Mulligan, F.J., and M.L. Doherty. 2008. Production diseases of the transition cow. Vet. J. 176:3-9.

Niozas, G., G. Tsousis, C. Malesios, I. Steinhöfel, C. Boscos, H. Bollwein, and M. Kaske. 2019a. Extended lactation in high-yielding dairy cows. II. Effects on milk production, udder health, and body measurements. J. Dairy Sci. 102: 811-823.

Niozas, G., G. Tsousis, I. Steinhöfel, C. Brozos, A. Römer, S. Wiedemann, H. Bollwein, and M. Kaske. 2019b. Extended lactation in high-yielding dairy cows. I. Effects on reproductive measurements. J. Dairy Sci. 102: 799-810.

Odensten, M.O., B. Berglund, K.P. Waller, and K. Holtenius. 2007. Metabolism and udder health at dry-off in cows of different breeds and production levels. J. Dairy Sci. 90:1417–1428.

Olori, V., S. Brotherstone, W. Hill, and B. McGuirk. 1997. Effect of gestation stage on milk yield and composition in Holstein Friesian dairy cattle. Livest. Prod. Sci. 52:167–176.

Opsomer, G., M. Coryn, H. Deluyker, and A. de Kruif. 1998. An analysis of ovarian dysfunction in high yielding dairy cows after calving based on progesterone profiles. Reprod. Domest. Anim. 33:193–204.

Opsomer, G., Y.T. Gröhn, J. Hertl, M. Coryn, H. Deluyker, and A. de Kruif. 2000. Risk factors for post partum ovarian dysfunction in high producing dairy cows in Belgium: a field study. Theriogenology 53:841-857.

Österman, S., and J. Bertilsson. 2003. Extended calving interval in combination with milking two or three times per day: Effects on milk production and milk composition. Livest. Prod. Sci. 82:139–149.

Österman, S., K. Östensson, K. Svennersten-Sjaunja, and J. Bertilsson. 2005. How does extended lactation in combination with different milking frequencies affect somatic cell counts in dairy cows? Livest. Prod. Sci. 96:225-232.

Peel, C.J., T.J. Fronk, D.E. Bauman, and R.C. Gorewit. 1983. Effect of exogenous growth hormone in early and late lactation on lactational performance of dairy cows. J. Dairy Sci. 66:776-782.

Penasa, M., M. de Marchi, and M. Cassandro. 2016. Effects of pregnancy on milk yield, composition traits, and coagulation properties of Holstein cows. J. Dairy Sci. 99:4864–4869.

Pinedo, P.J., A. Daniels, J. Shumaker, and A. De Vries. 2014. Dynamics of culling for Jersey, Holstein, and Jersey × Holstein crossbred cows in large multibreed dairy herds. J. Dairy Sci. 97:2886–2895.

Poppe, M., R.F. Veerkamp, M.L. van Pelt, and H.A. Mulder. 2020. Exploration of variance, autocorrelation, and skewness of deviations from lactation curves as resilience indicators for breeding. J. Dairy Sci. 103:1667–1684.

Pryce, J.E., M. Haile-Mariam, K. Verbyla, P.J. Bowman, M.E. Goddard, and B.J. Hayes. 2010. Genetic markers for lactation persistency in primiparous Australian dairy cows. J. Dairy Sci. 93:2202-2214.

Pushpakumara, P.G.A., N.H. Gardner, C.K. Reynolds, D.E. Beever, and D.C. Wathes. 2003. Relationships between transition period diet, metabolic parameters and fertility in lactating dairy cows. Theriogenology 60:1165-1185.

Rajala-Schultz, P.J., J.S. Hogan, and K.L. Smith. 2005. Short communication: Association between milk yield at dry-off and probability of intramammary infections at calving. J. Dairy Sci. 88:577–579.

Rastani, R.R., R.R. Grummer, S.J. Bertics, A. Gümen, M.C. Wiltbank, D.G. Mashek, and M.C. Schwab. 2005. Reducing dry period length to simplify feeding transition cows: Milk production, energy balance, and metabolic profiles. J. Dairy Sci. 88:1004-1014.

Rehn, H., B. Berglund, U. Emanuelson, G. Tengroth, and J. Philipsson. 2000. Milk production in Swedish dairy cows managed for calving intervals of 12 and 15 months. Acta Agric. Scan. A Anim. Sci. 50:263-271.

Rethmeier, J., J. Wenzlau, M. Wagner, S. Wiedemann, and L. Bachmann. 2019. Fertility parameters in German dairy herds: Associations with milk yield and herd size. Czech J. Anim. Sci. 64:459-464.

Roche, J.R. 2003. Effect of pregnancy on milk production and bodyweight from identical twin study. J. Dairy Sci. 86:777–783.

Roche, J.R., and D.P. Berry. 2006. Periparturient climatic, animal, and management factors influencing the incidence of milk fever in grazing systems. J. Dairy Sci. 89:2775–2783.

Roche, J.R., J.M. Lee, K.A. Macdonald, and D.P. Berry. 2007. Relationships among body condition score, body weight, and milk production variables in pasture-based dairy cows. J. Dairy Sci. 90:3802-3815.

Roche, J.R., N.C. Friggens, J.K. Kay, M.W. Fisher, K.J. Stafford, and D.P. Berry. 2009. Invited review: Body condition score and its association with dairy cow productivity, health, and welfare. J. Dairy Sci. 92:5769–5801.

Roche, J.R., J.K. Kay, N.C. Friggens, J.J. Loor, and D.P. Berry. 2013. Assessing and managing body condition score for the prevention of metabolic disease in dairy cows. Vet. Clin. N. Am. – Food Anim. Pract. 29:323-336.

Romero, C.J., E. Pine-Twaddell, D.I. Sima, R.S. Miller, L. He, F. Wondisford, and S. Radovick. 2012. Insulin-like growth factor 1 mediates negative feedback to somatotroph GH expression via POU1F1/CREB binding protein interactions. Mol. Cell. Biol. 32:4258-4269.

Rutten, C.J., W. Steeneveld, C. Inchaisri, and H. Hogeveen. 2014. An ex ante analysis on the use of activity meters for automated estrus detection: To invest or not to invest? J. Dairy Sci. 97:6869–6887.

Schindler, H., S. Eger, M. Davidson, D. Ochowski, E.C. Schermerhorn, and R.H. Foote. 1991. Factors affecting response of groups of dairy cows managed for different calving-conception intervals. Theriogenology 36:495-503.

Schmidt, G.H. 1989. Effect of length of calving intervals on income over feed and variable costs. J. Dairy Sci. 72:1605-1611.

Schneider, F., J.A. Shelford, R.G. Peterson, and L.J. Fisher. 1981. Effects of early and late breeding of dairy cows on reproduction and production in current and subsequent lactation. J. Dairy Sci. 64:1996-2002.

Schuh, K., H. Sadri, S. Häussler, L.A. Webb, C. Urh, M. Wagner, C. Koch, J. Frahm, S. Dänicke, G. Dusel, and H. Sauerwein. 2019. Comparison of performance and metabolism from late pregnancy to early lactation in dairy cows with elevated v. normal body condition at dry-off. Animal 13:1478-1488.

Sehested, J., C. Gaillard, J.O. Lehmann, G.M. Maciel, M. Vestergaard, M.R. Weisbjerg, L. Mogensen, L.B. Larsen, N.A. Poulsen, and T. Kristensen. 2019. Extended lactation in dairy cattle. Animal 13:65-74.

Shrestha, H.K., T. Nakao, T. Higaki, T. Suzuki, and M. Akita. 2004. Resumption of postpartum ovarian cyclicity in high-producing Holstein cows. Theriogenology 61:637–649.

Silvestre, A.M., A.M. Martins, V.A. Santos, M.M. Ginja, and J.A. Colaço. 2009. Lactation curves for milk, fat and protein in dairy cows: A full approach. Livest. Sci. 122:308–313.

Sørensen, J.T., and S. Østergaard. 2003. Economic consequences of postponed first insemination of cows in a dairy cattle herd. Livest. Prod. Sci. 79:145-153.

Sorensen, A., D.D. Muir, and C.H. Knight. 2001. Thrice-daily milking throughout lactation maintains epithelial integrity and thereby improves milk protein quality. J. Dairy Res. 68:15-25.

Sorensen, A., D.D. Muir, and C.H. Knight. 2008. Extended lactation in dairy cows: effects of milking frequency, calving season and nutrition on lactation persistency and milk quality. J. Dairy Res. 75:90-97.

Stangaferro, M.L., R. Wijma, M. Masello, M.J. Thomas, and J.O. Giordano. 2018. Economic performance of lactating dairy cows submitted for first service timed artificial insemination after a voluntary waiting period of 60 or 88 days. J. Dairy Sci. 101:7500–7516.

Steeneveld, W., and H. Hogeveen. 2012. Economic consequences of immediate or delayed insemination of a cow in oestrus. Vet. Rec. 171:17.

Strandberg, E., and C. Lundberg. 1991. A note on the estimation of environmental effects on lactation curves. Anim. Prod. 53: 399–402.

Strandberg, E., and P.A. Oltenacu. 1989. Economic consequences of different calving intervals. Acta Agric. Scan. A Anim. Sci. 39:407–420.

Tannenbaum, G.S., H.J. Guyda, and B.I. Posner. 1983. Insulin-like growth factors: a role in growth hormone negative feedback and body weight regulation via brain. Science 220:77-79.

Valizaheh, R., D.M. Veira, and M.A.G. von Keyserlingk. 2008. Behavioural responses by dairy cows provided two hays of contrasting quality at dry-off. Appl. Anim. Behav. Sci. 109:190-200.

Van Amburgh, M.E., D.M. Galton, D.E. Bauman, and R.W. Everett. 1997. Management and economics of extended calving intervals with use of bovine somatotropin. Livest. Prod. Sci. 50:15–28.

Van Dooren, N. 2019. The effects of extending the voluntary waiting period on services per conception in dairy cows and the added value of extended lactation for Dutch dairy farmers. MSc thesis, Wageningen University, Wageningen, the Netherlands.

Van Knegsel, A.T.M., H. van den Brand, J. Dijkstra, W.M. van Straalen, R. Jorritsma, S. Tamminga, and B. Kemp. 2007. Effect of glucogenic vs. lipogenic diets on energy balance, blood metabolites, and reproduction in primiparous and multiparous dairy cows in early lactation. J. Dairy Sci. 90:3397-3409.

Veerkamp, R.F., B. Beerda, and T. van der Lende. 2003. Effects of genetic selection for milk yield on energy balance, levels of hormones, and metabolites in lactating cattle, and possible links to reduced fertility. Livest. Prod. Sci. 83:257-275.

Vilar, M.J., and P.J. Rajala-Schultz. 2020. Dry-off and dairy cow udder health and welfare: Effects of different milk cessation methods. Vet. 262:105503.

Wall, E., M.P. Coffey, and G.E. Pollott. 2012. The effect of lactation length on greenhouse gas emissions from the national dairy herd. Animal 6:1857-1867.

Walsh, S.W., E.J. Williams, and A.C.O. Evans. 2011. A review of the causes of poor fertility in high milk producing dairy cows. Anim. Reprod. Sci. 123:127–138.

Wathes, D.C., V.J. Taylor, Z. Cheng, and G.E. Mann. 2003. Follicle growth, corpus luteum function and their effects on embryo development in postpartum dairy cows. Reprod. Suppl. 61:219-237.

Wathes, D.C., N. Bourne, Z. Cheng, G.E. Mann, V.J. Taylor, and M.P. Coffey. 2007. Multiple correlation analyses of metabolic and endocrine profiles with fertility in primiparous and multiparous cows. J. Dairy Sci. 90:1310-1325.

Weber, M.S., S. Purup, M. Vestergaard, R.M. Akers, and K. Sejrsen. 2000. Regulation of local synthesis of insulin-like growth factor-I and binding proteins in mammary tissue. J. Dairy Sci. 83:30-37.

Wilmink, J.B.M. 1987. Adjustment of test-da y milk, fat and protein yield for age, season and stage of lactation. Livest. Prod. Sci. 16:335–348.

Wilmink, J.B.M. 1988. Selection on fat and protein to maximise profit in dairy herds. Livest. Prod. Sci. 20:299-316.

Yart, L., V. Lollivier, L. Finot, J. Dupont, S. Wiart, M. Boutinaud, P.G. Marnet, and F. Dessauge. 2013. Changes in mammary secretory tissue during lactation in ovariectomized dairy cows. Steroids 78:973-981.

Zhao, W., X. Chen, J. Xiao, X.H. Chen, X.F. Zhang, T. Wang, Y.G. Zhen, and G.X. Qin. 2019. Prepartum body condition score affects milk yield, lipid metabolism, and oxidation status of Holstein cows. Asian-Australas. J. Anim. Sci. 32:1889.

Zobel, G., K. Leslie, D.M. Weary, and M.A.G. von Keyserlingk. 2013. Gradual cessation of milking reduces milk leakage and motivation to be milked in dairy cows at dry-off. J. Dairy Sci. 96:5064-5071.

Zobel, G., D.M. Weary, K.E. Leslie, and M.A.G. von Keyserlingk. 2015. Invited review: Cessation of lactation: Effects on animal welfare. J. Dairy Sci. 98:8263–8277.

Summary

In dairy farming, cows are usually managed to have a calf every year. A system with yearly calvings is expected to maximize yearly milk production due to the associated yearly peak in milk production in the beginning of every new lactation. In early lactation, dairy cows usually experience a negative energy balance (NEB) towards this peak in the first 8-10 weeks of lactation. This NEB is associated with impaired health and fertility. To reduce the frequency of periods with NEB, lactations could be deliberately extended by extending the voluntary waiting period for insemination (VWP). This increases the calving interval (CInt) and reduces the frequency of calvings. A lower frequency of calvings per cow results in fewer transitions related with calving on a herd level. However, a longer period in late lactation may be related with a lower milk output per year, and a risk for fattening at the end of the lactation with consequences for the subsequent transition period. The aim of this thesis was to evaluate the consequences of extended lactations for milk production, fertility, health, metabolism, and economic result. Additionally, the aim was to identify cow factors that determine the response of individual cows to an extended lactation.

To study this aim, a two-sided approach was applied: 1. A network of 14 Dutch dairy farmers that apply extended lactations on their farms was formed, to exchange knowledge and to evaluate data of these commercial farms, and 2. An animal experiment was conducted, where 154 cows in a randomized block design were assigned to a VWP of 50, 125, or 200 days, and monitored for a complete lactation and the first 6 weeks of the next lactation.

The main motivations of the dairy farmers in our network to extend the lactation on their farms were: fewer calving moments, fewer calves, drying off cows at a lower milk level, improved insemination rates, and less labor related with calving and calf rearing. In **chapter 2**, we investigated the consequences of extended lactations on these farms for fertility and milk production characteristics. It was expected that a delayed insemination would result in an improved reproductive performance, as milk production would be decreased, and cows could have recovered from the calving process and the NEB at the time of insemination. However, a greater calving to first service interval (CFSI) did not result in fewer inseminations or a higher conception rate. Possibly, the selection of high-producing cows by the farmers for a delayed insemination resulted in a similar milk production at the time of insemination, and as such a similar reproductive performance during that time. An increase in CInt was related with more services to conception and a lower conception rate at first service, which may indicate that the

longer CInt could be the result of impaired reproductive performance. Cows with the longest CInt had the greatest 305-d production, probably again partly due to selection by the farmers of the most productive cows for the longest lactation. These cows, however, did not have the greatest milk production per day of CInt, which may be explained by more days in late lactation (> 305 days) with a lower milk production when the CInt is longer. On most farms, peak production was lowest in the shortest CInt class, which may indicate a selection of low producing cows for a shorter CInt, or a better reproductive performance for cows with a lower milk production in early lactation. Lactation persistency increased with increasing CInt when the shortest CInt class was excluded, possibly due to a delayed effect of pregnancy on the lactation curve.

In **chapter 3**, we investigated the consequences of an extended VWP on milk production and persistency in an experimental setting with 154 cows on our dairy research farm. For both primiparous and multiparous cows, the VWP could be extended until 125 days without an effect on milk or fat- and protein-corrected milk (FPCM) production per day of CInt. When the VWP was extended until 200 days, milk production of multiparous cows was reduced compared with multiparous cows with shorter VWP. This may be attributed to more time in late lactation and relatively fewer days of peak production. For primiparous cows, an extension of the VWP until 200 days did not affect milk or FPCM production per day of CInt, possibly related to their greater lactation persistency compared with multiparous cows. In this study, the lactation persistency between day 100 in lactation and the start of dry-off was improved for cows with a VWP of 200 days compared with cows with a VWP of 50 days. Despite this improvement in lactation persistency, cows with an extended VWP had a lower milk production before dry-off compared with cows with a VWP of 50 days. This lower milk production before dry-off may be beneficial for udder health. However, this lower milk production at the end of the lactation may be related with fattening of cows during that time. Indeed, multiparous but not primiparous cows with a VWP of 200 days had a greater body condition at the end of their extended lactation.

In **chapter 4**, we investigated the consequences of an extended VWP on metabolism during different phases of the lactation in the same experiment. An extended VWP implies inseminating cows later in lactation, when milk production can be expected to be reduced. Indeed, around the end of the VWP, the FPCM production was lowest for multiparous cows with a VWP of 200 days, intermediate for multiparous cows with a VWP of 125, and greatest for multiparous cows with a VWP of 50 days. Moreover, around the end of the VWP, multiparous cows with a VWP of 200 days had a greater plasma insulin and insulin-like growth factor 1 (IGF-1) concentration and partitioned less energy to milk than to maintenance compared with multiparous cows with a shorter VWP. For primiparous cows, the VWP did not affect milk production or metabolism around the end of the VWP. When insemination is delayed, pregnancy takes place at a later stage in lactation. During pregnancy, multiparous cows with a VWP of 200 days had a lower FPCM production, a greater plasma concentration of insulin and IGF-1, a greater body condition score, and these cows gained more body weight and partitioned less energy to milk than to maintenance compared with multiparous cows with a shorter VWP. Primiparous cows with a VWP of 125 days had a greater plasma insulin concentration compared with primiparous cows with a VWP of 50 days during pregnancy, but the VWP did not affect milk production or body condition for primiparous cows during pregnancy. Possibly related with these limited effects of the VWP on metabolism of primiparous cows during pregnancy, the VWP did not affect milk production or metabolism during the start of the next lactation for primiparous cows. For multiparous cows, a VWP of 200 days resulted in a greater body condition score during the start of the next lactation, a more severe NEB, and a greater plasma non-esterified fatty acid concentration compared with multiparous cows with a VWP of 50 days.

This indicates that, for multiparous cows, an extended lactation may increase the risk for fattening at the end of the extended lactation, and a more severe NEB during the start of the subsequent lactation. A more severe NEB during the start of the lactation could be related with an increased risk for diseases. However, not all multiparous cows gained body condition at the end of the extended lactation or had a severe NEB during the start of the next lactation. Variation among individual cows in for example milk production and energy partitioning could affect the performance in their lactation. Independent of the VWP, a higher milk production and a lower body condition before insemination were associated with a higher milk production and a lower body condition score at the end of the lactation. As such, multiparous cows with a high milk production and a low body condition may be more suitable for an extended lactation, to limit the risk for fattening at the end of the extended lactation.

In **chapter 5**, we investigated the consequences of an extended VWP on economic performance based on the results of the same experiment. In that study, cows with a VWP of 200 days had lower total yearly revenues compared with cows with a VWP of 50 days, mainly due to lower yearly revenues from milk. However, cows with a VWP of 200 days also had lower total yearly costs compared with cows with a VWP of 50 days, mainly due to lower yearly costs for concentrate supply and inseminations. Numerically, cows with a VWP of 200 days also had lower yearly costs for veterinary treatments and culling. Due to this partial compensation of the lower revenues by the lower costs, the yearly net partial cashflow (NPCF) was not significantly associated with VWP. However, numerically, the yearly NPCF was €47 lower for cows with a VWP of 125 and €102 lower for cows with a VWP of 200 days compared with cows with a VWP of 50 days. One explanation for the absence of a significant difference among the 3 VWP groups is the large variation in NPCF among individual cows. To investigate this variation, cows were divided over 3 economic classes based on their yearly NPCF. Cows with a yearly NPCF of > €1,400/year had a greater milk production, protein production, fat production, and lactose production per day of CInt compared with cows with a yearly NPCF of < €1,100/year. Moreover, cows with the greater NPCF had on average 5 fewer veterinary treatments during the experiment. The economic class was not related with the VWP, CFSI, or CInt.

In **chapter 6**, the results of this thesis from both the data of the commercial dairy farms and the experiment were compared with each other and with earlier literature on extended lactations. It is important to distinguish the terms VWP, CFSI, and CInt, as the VWP may be a management strategy, and the CFSI and CInt are affected by estrus detection and reproductive performance. When cows were selected for an extended CInt until 531 days in practice, on most farms milk production per day of CInt was not reduced for these cows. In experimental settings, where there was no selection of specific cows for an extended VWP, milk production often decreased for multiparous cows. In addition, in experimental settings often reproductive performance improved with an extension of the VWP, while in practice this was not the case, possibly as a result of high-producing cows being selected for a delayed insemination.

To conclude, this thesis assessed the consequences of an extended lactation, both on commercial farms and in an experimental setting. Primiparous cows can handle an extended lactation very well, as an extended VWP did not affect their milk production or metabolism. For multiparous cows, a customized lactation length may limit the risk for a low milk production and fattening at the end of the extended lactation. When suitable cows are selected, extended lactations may contribute to the sustainability of dairy farming, as it results in a lower frequency of critical calving moments, fewer surplus calves, and less labor associated with calving cows and rearing of calves.

Samenvatting

De meeste melkveehouders streven naar een tussenkalftijd (TKT) van een jaar. Dit jaarlijks afkalven is gericht op het maximaliseren van de melkproductie, door de piek in melkproductie aan het begin van iedere nieuwe lactatie. In de eerste 8 – 10 weken van een nieuwe lactatie hebben melkkoeien vaak een negatieve energiebalans (NEB). Deze NEB is geassocieerd met een hogere ziekte-incidentie en een verminderde vruchtbaarheid. Om de frequentie van deze perioden met een NEB te verminderen zouden lactaties bewust verlengd kunnen worden door de vrijwillige wachtperiode tot inseminatie (VWP) te verlengen. Dit verlengt de tussenkalftijd (TKT) en vermindert de frequentie van afkalven. Een lagere frequentie van afkalven resulteert op koppelniveau in minder transities rondom afkalven. Een langere periode laat in lactatie kan echter samenhangen met een lagere melkproductie per jaar en meer risico op vervetting aan het eind van de lactatie met consequenties voor de volgende transitieperiode rondom het afkalven. Het doel van deze thesis was om de gevolgen van het verlengen van de lactatie voor melkproductie, vruchtbaarheid, gezondheid, stofwisseling, en economisch resultaat te onderzoeken. Daarnaast was het doel koefactoren te identificeren die de respons van individuele koeien op een verlengde lactatie bepalen.

Dit doel is onderzocht met een tweezijdige benadering. 1. Er is een netwerk gevormd van 14 Nederlandse melkveehouders die bewust de lactatie van (een deel van) hun koppel verlengen, om kennis uit te wisselen en gegevens van deze commerciële bedrijven te analyseren, en 2. Er is een dierproef uitgevoerd, waarin 154 koeien in een gerandomiseerd blok design toegewezen zijn aan een VWP van 50, 125, of 200 dagen, en gevolgd zijn voor een volledige lactatie en de eerste 6 weken van de volgende lactatie.

De belangrijkste motivaties van de melkveehouders in ons netwerk om de lactatielengte op hun bedrijf te verlengen waren: minder afkalfmomenten, minder kalveren, koeien droogzetten bij een lager melkniveau, betere vruchtbaarheidsresultaten, en minder arbeid gerelateerd aan het afkalven van koeien en het verzorgen van kalveren. In **hoofdstuk 2** onderzochten we de gevolgen van de verlengde lactaties op deze bedrijven voor vruchtbaarheidsresultaten en melkproductiekenmerken. De verwachting was dat een uitgestelde inseminatie zou leiden tot betere vruchtbaarheidsresultaten, omdat de melkproductie later in lactatie vaak afgenomen is, en koeien de kans hebben gehad om te herstellen van het afkalven en de NEB. Een groter interval tussen afkalven en eerste inseminatie (IAI) resulteerde echter niet in minder inseminaties tot dracht of hogere bevruchtingspercentages. Mogelijk heeft de selectie door de

melkveehouder van hoogproductieve koeien voor een uitgestelde inseminatie geleid tot een vergelijkbare melkproductie op het moment van inseminatie en daarmee vergelijkbare vruchtbaarheidsresultaten. Een langere TKT was gerelateerd aan meer inseminaties tot dracht en een lager bevruchtingspercentage bij eerste inseminatie, wat erop kan wijzen dat een langere TKT het gevolg kan zijn van verminderde vruchtbaarheid. Koeien met de langste TKT hadden de hoogste 305-d productie, waarschijnlijk wederom mede door selectie van de meest productieve koeien voor de langste lactatie door de melkveehouder. Deze koeien hadden echter niet de hoogste melkproductie per dag TKT, wat kan worden verklaard door meer dagen laat in de lactatie (> 305 dagen) met een lagere melkproductie wanneer de TKT verlengd is. Op de meeste bedrijven was de piekproductie het laagst in de kortste TKT-klasse, wat kan duiden op een selectie van laagproductieve koeien voor een kortere TKT, of op betere vruchtbaarheidsresultaten voor koeien met een lagere melkproductie in het begin van de lactatie. De lactatiepersistentie nam toe met toenemende TKT wanneer de kortste TKT-klasse niet werd meegenomen, mogelijk als gevolg van een later effect van dracht op de lactatiecurve.

In **hoofdstuk 3** onderzochten we de gevolgen van een verlengde VWP voor de melkproductie en lactatiepersistentie in een experimentele setting met 154 koeien op ons melkveeproefbedrijf. Voor zowel eerstekalfs- als meerderekalfskoeien kon de VWP verlengd worden tot 125 dagen zonder effect op de melkproductie of op de vet- en eiwit-gecorrigeerde melkproductie. Wanneer de VWP verlengd werd tot 200 dagen hadden meerderekalfskoeien een lagere melkproductie vergeleken met meerderekalfskoeien met een kortere VWP. Dit kan worden toegeschreven aan meer tijd in de late lactatie en een kleiner aandeel van de piek ten opzichte van de complete lactatie. Voor eerstekalfskoeien had een verlenging van de VWP tot 200 dagen geen effect op de melkproductie per dag TKT, mogelijk gerelateerd aan hun hogere lactatiepersistentie in vergelijking met meerderekalfskoeien. De lactatiepersistentie tussen dag 100 in lactatie en het begin van de droogstand was hoger voor koeien met een VWP van 200 dagen vergeleken met koeien met een VWP van 50 dagen. Ondanks deze verbetering in lactatiepersistentie hadden koeien met een langere VWP een lagere melkproductie kort voor de droogstand. Deze lagere melkproductie voor het droogzetten kan gunstig zijn voor de uiergezondheid, maar kan ook een risico zijn voor het vervetten van koeien aan het eind van de lactatie. Meerderekalfskoeien met een VWP van 200 dagen hadden inderdaad een hogere lichaamsconditiescore aan het eind van hun verlengde lactatie vergeleken met meerderekalfskoeien met kortere VWP. Voor eerstekalfskoeien was dit effect er niet.

In **hoofdstuk 4** onderzochten we de gevolgen van een verlengde VWP op de stofwisseling tijdens verschillende fasen van de lactatie in hetzelfde experiment. Een verlengde VWP impliceert dat koeien later in de lactatie geïnsemineerd worden, wanneer verwacht kan worden dat de melkproductie dan afgenomen is. Inderdaad, rond het einde van de VWP was de melkproductie het laagst voor meerderekalfskoeien met een VWP van 200 dagen, gemiddeld voor meerderekalfskoeien met een VWP van 125 dagen, en het hoogst voor meerderekalfskoeien met een VWP van 50 dagen. Bovendien hadden meerderekalfskoeien met een VWP van 200 dagen rondom het einde van de VWP een hogere plasma insuline en IGF-1 concentratie en besteedden deze koeien relatief minder energie aan melk ten opzichte van onderhoud vergeleken met meerderekalfskoeien met een kortere VWP. Voor eerstekalfskoeien had de VWP geen invloed op de melkproductie of de stofwisseling rondom het einde van de VWP. Wanneer de inseminatie wordt uitgesteld, zijn koeien drachtig op een later moment in de lactatie. Tijdens de dracht hadden meerderekalfskoeien met een VWP van 200 dagen een lagere melkproductie, een hogere plasmaconcentratie van insuline en IGF-1, een hogere lichaamsconditiescore, en bovendien hadden deze koeien een hogere toename in lichaamsgewicht en besteedden ze relatief minder energie aan melk ten opzichte van onderhoud vergeleken met meerderekalfskoeien met een kortere VWP. Eerstekalfskoeien met een VWP van 125 dagen hadden een hogere plasma insulineconcentratie vergeleken met eerstekalfskoeien met een VWP van 50 dagen, maar de lengte van de VWP had geen effect op de melkproductie of lichaamsconditie van eerstekalfskoeien tijdens de dracht. Ook had de VWP geen effect op de melkproductie of de stofwisseling tijdens de start van de volgende lactatie voor eerstekalfskoeien, mogelijk gerelateerd aan de beperkte effecten van de VWP op de melkproductie, stofwisseling, en groei van eerstekalfskoeien tijdens de dracht. Bij meerderekalfskoeien resulteerde een VWP van 200 dagen in een diepere NEB aan het begin van de volgende lactatie, een hogere lichaamsconditiescore, en een hogere plasmaconcentratie van vrije vetzuren vergeleken met meerderekalfskoeien met een VWP van 50 dagen.

Deze resultaten geven aan dat een verlengde lactatie voor meerderekalfskoeien het risico op vervetting aan het eind van de lactatie kan verhogen, en daarmee het risico op een diepere NEB gedurende de start van de volgende lactatie. Een diepere NEB kan gerelateerd zijn aan een verhoogd risico op ziekte. Niet alle meerderekalfskoeien hadden echter een hogere lichaamsconditie aan het eind van hun verlengde lactatie, of een diepere NEB in het begin van de volgende lactatie. De variatie tussen individuele koeien in bijvoorbeeld melkproductie en energieverdeling beïnvloedt de prestatie in hun lactatie. Onafhankelijk van de lengte van de

VWP waren een hogere melkproductie en een lagere lichaamsconditie kort voor inseminatie geassocieerd met een hogere melkproductie en een lagere lichaamsconditiescore aan het eind van de lactatie. Zo zouden meerderekalfskoeien met een hogere melkproductie en een lagere lichaamsconditie meer geschikt kunnen zijn voor een verlengde lactatie, omdat zo het risico op vervetting aan het eind van de lactatie beperkt kan worden.

In **hoofdstuk 5** onderzochten we de gevolgen van een verlengde VWP op de economische prestatie op basis van de resultaten van hetzelfde experiment. Koeien met een VWP van 200 dagen hadden lagere totale jaarlijkse opbrengsten vergeleken met koeien met een VWP van 50 dagen, voornamelijk door lagere jaarlijkse melkopbrengsten. Koeien met een VWP van 200 dagen hadden echter ook lagere totale jaarlijkse kosten vergeleken met koeien met een VWP van 50 dagen, voornamelijk door lagere jaarlijkse kosten voor krachtvoer en inseminaties. Numeriek gezien hadden koeien met een VWP van 200 dagen ook lagere jaarlijkse kosten voor veterinaire behandelingen en afvoer. Door deze gedeeltelijke compensatie van de lagere opbrengsten door de lagere kosten was de jaarlijkse netto kasstroom niet significant geassocieerd met de VWP. Numeriek was de jaarlijkse netto kasstroom echter € 47 lager voor koeien met een VWP van 125 dagen, en € 102 lager voor koeien met een VWP van 200 dagen vergeleken met koeien met een VWP van 50 dagen. Een verklaring voor het ontbreken van een significant verschil tussen de 3 VWP-groepen is de grote variatie in kasstroom tussen individuele koeien. Om deze variatie te onderzoeken hebben we koeien verdeeld over 3 economische klassen op basis van hun jaarlijkse netto kasstroom. Koeien met een jaarlijkse kasstroom van > €1,400/jaar hadden een hogere melkproductie, eiwitproductie, vetproductie en lactoseproductie per dag TKT vergeleken met koeien met een jaarlijkse kasstroom van < €1,100/jaar. Bovendien kregen koeien met hogere kasstroom gemiddeld 5 veterinaire behandelingen minder tijdens het experiment. De economische klasse was niet gerelateerd aan de VWP, IAI of TKT.

In **hoofdstuk 6** zijn de resultaten van dit proefschrift van zowel de commerciële melkveebedrijven als het experiment vergeleken met elkaar en met eerdere literatuur over verlengde lactaties. Het is belangrijk om onderscheid te maken tussen de termen VWP, IAI en TKT, aangezien de VWP een managementstrategie kan zijn en de IAI en TKT worden beïnvloed door tochtdetectie en vruchtbaarheidsresultaten. Wanneer koeien in de praktijk werden geselecteerd voor een verlengde TKT tot 531 dagen was op de meeste bedrijven de melkproductie per dag TKT van deze koeien niet verlaagd. In experimenten, waar geen specifieke koeien werden geselecteerd voor een verlengde VWP, nam de melkproductie per dag TKT vaak af voor meerderekalfskoeien. Bovendien verbeterden de reproductieprestaties vaak met een verlenging van de VWP in experimenten, terwijl dit in de praktijk niet het geval was, mogelijk als gevolg van het selecteren van hoogproductieve koeien voor een uitgestelde inseminatie.

In conclusie zijn in dit proefschrift de gevolgen van een verlengde lactatie onderzocht, zowel op commerciële melkveebedrijven als in een experimentele setting. Eerstekalfskoeien kunnen een verlengde lactatie heel goed aan, aangezien een verlengde VWP geen invloed had op hun melkproductie of stofwisseling. Voor meerderekalfskoeien kan een lactatielengte op maat het risico op een lage melkproductie en vervetting aan het einde van de verlengde lactatie beperken. Wanneer geschikte koeien worden geselecteerd, kan een langere lactatie bijdragen aan de verduurzaming van de melkveehouderij, omdat dit resulteert in een lagere frequentie van kritische afkalfmomenten, minder overtollige kalveren, en minder arbeid gerelateerd aan het afkalven van koeien en het verzorgen van kalveren.

Dankwoord/ acknowledgements

De afgelopen 4 (nou ja, 4,5/ 5) jaar heb ik met veel plezier gewerkt aan mijn proefschrift, waarbij de hulp, ideeën, en samenwerkingen van anderen onmisbaar zijn geweest. Daarom wil ik bij deze iedereen die in de afgelopen tijd een bijdrage heeft geleverd graag bedanken!

Als eerste wil ik graag mijn begeleiders bedanken. Voorop: Ariëtte en Roselinde, het was superfijn met jullie samen te werken! Bedankt voor de motiverende en gezellige overleggen waarbij naast de inhoud ook de sociale kant aandacht kon krijgen. Ariëtte, bedankt voor je heldere schrijfinzichten, je netwerktips, en je nuance. Roselinde, bedankt voor je probleemoplossend vermogen, je praktische en nuchtere instelling, en vooral ook voor de hulp bij Dairy Campus en de dataset in het begin. Ook wil ik graag mijn 2 promotoren bedanken. Bas, bedankt voor je scherpe opmerkingen bij mijn papers (ook al was het soms wat ontcijferwerk), de discussies over fysiologie, en je talent om knopen door te hakken. Henk, ik vond het in het begin behoorlijk spannend om iets met economie te gaan doen – nogal een ver van mijn bed show. Jij hield me scherp als ik hier te kort door de bocht ging. Bedankt voor je expertise en enthousiasme!

Naast deze begeleiders heb ik ook een soort officieuze begeleider gehad: Akke, bedankt dat je altijd klaarstond om me te helpen en al mijn vragen te beantwoorden! Door je positieve instelling ben je een echte steun voor me geweest de afgelopen jaren. Naast deze fijne samenwerking is het ook altijd heel gezellig met je – bedankt voor de leuke etentjes, filmavonden, en wandelingen!

The experiment was also part of the thesis of another PhD student. Junnan, thank you for being my colleague during our project! I was really happy that you started only half a year after me, so that we could experience everything together. I enjoyed going to Dairy Campus together every week (happy that my car was a good bed for you so now and then when I picked you up at 5.30 in the morning :-P), taking samples of the cows, walking around the farm together with Jan and our wheelbarrow, and spending the rest of the day in the lab to centrifuge, pipet, and sort all the plasma and milk samples. I really enjoyed these (sometimes very long) days!

Het experiment dat onderdeel was van mijn PhD begon eind 2017 en eindigde in januari 2020 – een hele tijd, en hier hebben dan ook veel mensen aan meegeholpen. Fedde en Hester, bedankt voor alle hulp bij de proef en de werklijsten! Fedde, ik kon altijd bij je aankloppen als iets uit de data me niet helemaal duidelijk was. Daarnaast ook grote dank voor alle dierverzorgers en medewerkers van Dairy Campus! Jan, je hebt van het begin tot (bijna) het eind van de proef iedere week geholpen met het nemen van de bloedmonsters, enorm bedankt! En dat is nogal een werk, iedere week zo'n 80 koeien verspreid over Dairy Campus zien te vinden. Het was gezellig en leerzaam met je samen te werken. Hans, bedankt voor alle hulp tijdens de proef, en bij het bloedprikken in het staartje van de proef. Ebele, bedankt voor je hulp, en voor je aandacht voor de koeien en de proef. Gerard, bedankt voor je goede oog op de koeien en het in de gaten houden van hun welzijn!

Ik heb ze niet geteld, maar het moet ongeveer neerkomen op 60.000 buisjes met serum/ plasma en 28.000 buisjes met melk. Al deze monsters moesten niet alleen verzameld worden, maar ook opgeslagen en geanalyseerd. Over de opslag: Ger, bedankt voor je hulp bij de vriescel op Carus. Er was nogal eens wat aan de hand, en jij stond altijd klaar om de telefoon op te nemen en te helpen. Tamme, bedankt voor je snelle actie toen de vriezer de hitte écht niet meer aankon en je er maar een tuinslang op gezet hebt. Over de analyses: Rudie, bedankt voor de (snelle en vrij last-minute) analyse op het RIA lab bij ADP! Het was misschien niet het originele plan, maar het was heel fijn dat we met al deze monsters bij jou terecht konden en dat je zo veel mee kon denken. Joop, bedankt voor je hulp bij al mijn vragen rondom het lab, en ook voor de analyse van de biestmonsters. Rupert and Josef, thank you for all the analyses you did in Bern! And all that only for some Milka... ;-). Not only did you analyze so many samples, but I also enjoyed the discussions that we had on the results, your valuable input on the interpretation of the data, and your kind and productive attitudes. Thank you for the very nice collaboration!

Ook het netwerk van melkveehouders ben ik zeer dankbaar! Voor de data die ik van jullie heb mogen ontvangen en analyseren, voor de enquêtes die jullie (meerdere keren) wilden invullen, en vooral ook voor de vele bijeenkomsten die we samen gehad hebben waarbij jullie input zo waardevol was. Zelf kom ik niet van een melkveebedrijf, en vooral tijdens deze bijeenkomsten heb ik ontzettend veel geleerd over de praktische kant van de melkveehouderij, en over duurmelken in het bijzonder. Bedankt voor jullie enthousiaste bijdrage, waarin jullie motivaties, verschillende strategieën en ideeën daarbij uitvoerig besproken konden worden!

Natuurlijk ook bedankt aan de financiers van het project, ZuivelNL en het Ministerie van Landbouw, Natuur en Voedselkwaliteit, en aan de stuurgroep 'Lactatie op Maat': dankzij jullie kon dit project uitgevoerd worden. Hiernaast wil ik ook CRV bedanken voor de levering van de data en de bijdrage aan de bijeenkomsten met het netwerk en de werkgroep.

Lora en Nanette, bedankt voor jullie altijd open deur en alle hulp tijdens mijn PhD! Jullie hebben overal antwoord op en zijn altijd bereid om te helpen, dat heb ik erg gewaardeerd.

Studenten die hebben bijgedragen in de vorm van interviews met de veehouders, analyses van progesteron en biest in het lab, data-analyse, of andere projecten: Neeltje, Hanneke, Annemieke, Anne van Heesbeen, Anne Hoogeveen, Danniek, Noëlle, Joas, en Danie – bedankt voor jullie enthousiasme voor dit onderwerp en de fijne samenwerkingen!

De informele bijdragen in de afgelopen tijd worden zeker ook gewaardeerd. People from the tropical room, thank you for the nice time at ADP! And special thanks to the ADP-cow-people, formerly partly known as the WHYDRY team ;-). Novi, Renny, Wei, Akke, Junnan, Binyameen, Xiaodan, Yapin, and Ariette, I had a lot of fun with you: nice coffees, pancake dinners, farm visits, and most of all the cycling trips to Herveld or Ede. Ariette, thank you for welcoming all of us in your house which always feels like a holiday!

Daarnaast wil ik graag mijn vrienden bedanken! Vriendjes van de PLUSboot: bedankt voor de zeiltripjes, skitripjes, lange avonden, weekendjes weg, onzindiscussies, en goeie slechte grappen. Ik vind het superleuk dat zoveel van jullie nog in (de buurt van) Wageningen wonen, en we elkaar dus regelmatig en makkelijk op kunnen zoeken. PLUS geeft meer – véél meer! Naast Wageningen ben ik zo af en toe nog rondom Eindhoven te vinden: Team Eindhoven, superfijn dat we nog zo ongeveer dezelfde middelbare-school-vriendengroep zijn. Ook al zit er voor mij soms lang tussen, het voelt alsof we altijd weer door kunnen gaan waar we gebleven waren. Iris, bij jou kan ik altijd terecht, bedankt voor alle fijne gesprekken en wandelingen! Carmen, toen ik pas in Wageningen kwam werd je mijn huisgenootje, en dat mochten we zo'n 6 jaar volhouden. Bedankt voor alle gezellige wijnavonden (tot het oor!), fijne gesprekken, gekke avonturen, en leuke uitstapjes, en ook dat je Puck in huis bracht en ik daar 2 dagen per week op mocht passen!

Ook bedankt aan Ramon, Leoni, Vincent en Wouter. Ik voel me altijd heel welkom bij jullie en overal bij betrokken – dat waardeer ik!

Ten slotte: Jan en Arlette, ik ben blij dat we zo'n hecht gezin zijn en kan met alles altijd bij jullie terecht. Bedankt voor al jullie hulp en steun, dat ik altijd welkom ben, en voor de gezelligheid, de lol, en de leuke dingen die we samen delen. Laurens, in de afgelopen 4,5 jaar zijn we gaan samenwonen, hebben we een huis gekocht, en halverwege mijn PhD werd je ook nog mijn nieuwe thuiswerkcollega. Ik vond het een erg bijzondere en leuke tijd. Ik heb heel veel zin in alles wat we nog samen gaan beleven!

About the author

Eline Burgers was born on July 2nd, 1991, in Valkenswaard, the Netherlands. After completing her BSc in Animal Sciences at Wageningen University in 2015, she continued with her MSc in Animal Sciences. She specialized in Animal Health and Behaviour, at the chair groups Adaptation Physiology and Animal Production Systems. During her thesis at Adaptation Physiology, she studied health and performance of piglets in a group lactation system. During her thesis at Animal Production Systems, she studied the preference of dairy cows for an outdoor open pack or a freestall barn during winter. This thesis was carried out in collaboration with the University of British Columbia, and to perform the research Eline spent 6 months living at the UBC Dairy Education and Research Centre in Agassiz, Canada.

After graduation, Eline started with her PhD in the Adaptation Physiology Group and the Animal Nutrition department at Wageningen University & Research. In collaboration with the Business Economics Group, she investigated consequences of an extended lactation for milk production, lactation persistency, fertility, health, metabolism, and economic performance of individual dairy cows. Her PhD was part of an interdisciplinary project called 'Customized lactation length', which was financed by DairyNL (ZuivelNL; organization of the Dutch dairy supply chain) and the Dutch Ministry of Agriculture, Nature and Food Quality (LNV) as part of the research program One Health for Food (1H4F).

Since May 2022, Eline is working as a researcher at the Animal Nutrition department of Wageningen Livestock Research.

Publications

Refereed scientific journals

Burgers, E.E.A., A. Kok, R.M.A. Goselink, H. Hogeveen, B. Kemp, and A.T.M. van Knegsel. 2022. Revenues and costs of dairy cows with different voluntary waiting periods based on data of a randomized control trial. J. Dairy Sci. 105: 4171-4188.

Burgers, E.E.A., A. Kok, R.M.A. Goselink, H. Hogeveen, B. Kemp, and A.T.M. van Knegsel. 2021. Effects of extended voluntary waiting period from calving until first insemination on body condition, milk yield, and lactation persistency. J. Dairy Sci. 104: 8009-8022. (Editor's Choice)

Burgers, E.E.A., A. Kok, R.M.A. Goselink, H. Hogeveen, B. Kemp, and A.T.M. van Knegsel. 2021. Fertility and milk production on commercial dairy farms with customized lactation lengths. J. Dairy Sci. 104: 443-458.

Smid, A. M. C., E.E.A. Burgers, D.M. Weary, E.A.M. Bokkers, and M.A.G. von Keyserlingk. 2019. Dairy cow preference for access to an outdoor pack in summer and winter. J. Dairy Sci. 102: 1551-1558.

Ma, J., E.E.A. Burgers, A. Kok, R.M.A. Goselink, T.J.G.M. Lam, B. Kemp, and A.T.M. van Knegsel. 2022. Consequences of extending the voluntary waiting period for insemination on reproductive performance in dairy cows. Anim. Reprod. Sci. 244. *In press.*

Van Knegsel, A.T.M., E.E.A. Burgers, J. Ma, R.M.A. Goselink, and A. Kok. 2022. Extending Lactation Length: Consequences for Cow, Calf and Farmer. J. Anim. Sci. *In press.*

Burgers, E.E.A., R.M.A. Goselink, R.M. Bruckmaier, J.J. Gross, R. Jorritsma, B. Kemp, A. Kok, and A.T.M. van Knegsel. Effect of voluntary waiting period on metabolism of dairy cows during different phases of the lactation. *Submitted.*

Abstracts in conference proceedings

Burgers, E.E.A., A. Kok, R.M.A. Goselink, and A.T.M. van Knegsel. 2019. Customising lactation length: impact of calving interval, parity, and lactation persistency on milk production of dairy cows. In: Proceedings of the 17th International Conference on Production Diseases in Farm Animals, 27-29 June, Bern, Switzerland – p. 148.

Burgers, E.E.A., R.M.A. Goselink, B. Kemp, and A.T.M. van Knegsel. 2020. Reducing the number of critical transitions for dairy cows: effects on milk yield, body weight, and body condition score. In: WIAS Annual Conference 2020: Frontiers in Animal Sciences, 13-14 February, Lunteren, the Netherlands – p. 60.

Burgers, E.E.A., R.M.A. Goselink, B. Kemp, and A.T.M. van Knegsel. 2020. Reducing the number of critical transitions for dairy cows: effects on milk yield and health. In: Book

of Abstracts of the 71st Annual Meeting of the European Federation of Animal Science, 1-4 December, Virtual Meeting – p. 226.

Burgers, E.E.A., A. Kok, R.M.A. Goselink, H. Hogeveen, B. Kemp, and A.T.M. van Knegsel. 2021. Revenues and costs of dairy cows with different voluntary waiting periods until first insemination. In: WIAS Annual Conference 2021: Resilience, 28-29 April, Online COVID-friendly edition, – p. 20.

Burgers, E.E.A., A. Kok, R.M.A. Goselink, H. Hogeveen, B. Kemp, and A.T.M. van Knegsel. 2021. Net partial cashflow of dairy cows with different voluntary waiting periods until first insemination. In: Abstracts of the 2021 American Dairy Science Association Annual Meeting, J. of Dairy Sci. 104, No. Supplement 1, 11-14 July, Virtual Meeting – p. 52.

Burgers, E.E.A., R.M.A. Goselink, R.M. Bruckmaier, J.J. Gross, R. Jorritsma, B. Kemp, A. Kok, A.H. van Ruitenbeek, and A.T.M. van Knegsel. 2022. Effect of voluntary waiting period on metabolism of dairy cows in different stages of lactation. In: 27th WIAS Annual Conference 2022: Collective Action, 11 February, Lunteren, the Netherlands – p. 36.

Other publications related to this thesis

Burgers, E.E.A., R.M.A. Goselink, and A.T.M. van Knegsel. 2022. Lactatie op maat: verlengen van de lactatie van melkvee: effecten op melkproductie, gezondheid en vruchtbaarheid van de koe en het economisch resultaat. Wageningen University & Research, Wageningen, Nederland. 112 blz.

Enter, M. 2022. Melkveehouderij: lactatie valt te verlengen/ Dairy farming: lactation can be extended. Resource 16, 30-4-2022: p. 9.

Purmer, M. 2021. Duurmelken: de economie regeert. Boerderij 107, 26-10-2021: p. 3.

Purmer, M. 2021. Vroeg afkalven en dan langer doormelken. Boerderij 107, 26-10-2021: p. 16 – 17.

Bloemberg-Van der Hulst, M. 2021. Melkveehouder Van den Bosch: ‘Elk jaar kalf niet meer van deze tijd’. Nieuwe Oogst, 9-1-2021: p. 16.

De Vries, G. 2019. Duurmelken voor minder trammelant. Veeteelt, 1-6-2019: p. 28-29.

Kwekkeboom, M. 2018. Opinie: Tussenkalftijd is te beperkt kengetal. Boerderij 103, 1-8-2018: p. 55.

Pellikaan, F. 2018. Korte tussenkalftijd verliest heel langzaam fans. Veeteelt, 1-3-2018: p. 36 – 41.

Education certificate

Completed training and supervision plan[1]

The basic package (3.0 ECTS)	
WIAS Introduction Day	2017
Research integrity & ethics in animal sciences	2017
Course on essential skills	2017

Disciplinary competences (15.8 ECTS)	
Writing PhD proposal	2017-2018
Animal health and immunology discussion group	2017-2018
Introduction to laboratory animal science course	2018
Species-specific laboratory animal science course “ruminants”	2018
Advanced statistics course: design of experiments	2019

Professional competences (7.7 ECTS)	
Brain training	2018
Project and time management	2018
Presenting with impact	2019
Mobilising your scientific network	2019
Scientific writing	2020
Career orientation	2021
Last stretch of the PhD and writing propositions	2021
The final touch: writing the general introduction and discussion	2021

Societal relevance (1.5 ECTS)	
Societal impact of your research	2020

Presentation skills (maximum of 4.0 ECTS)	
BASF workshop, Merelbeke, Belgium (oral)	2018
ICPD, Bern, Switzerland (oral)	2019
WIAS Annual Conference, Lunteren, the Netherlands (poster)	2020
EAAP, virtual meeting (oral)	2020
WIAS Annual Conference, virtual meeting (oral)	2021
ADSA, virtual meeting (oral)	2021
WIAS Annual Conference, Lunteren, the Netherlands (oral)	2022

Other scientific meetings (3.6 ECTS)	
Organizing meetings with farmers of the customized lactation length network + oral presentations, the Netherlands	2017-2022
International dairy nutrition symposium 'new perspectives on transition cow management', Wageningen, the Netherlands	2017
Discover conference – effects of stress on health and production in dairy cows, Chicago, USA	2018
Nutrition in transition - Schothorst Feed Research, Nijkerk, the Netherlands	2019

Teaching competences (maximum of 6.0 ECTS)	
Supervision of 3 BSc thesis students	2020-2022
Supervision of 4 MSc thesis students	2018-2022
Supervision student groups ‘Introduction to Animal Sciences’	2018, 2019
Supervision student group ‘Adaptation Physiology’	2021

Extra tasks (8.0 ECTS)	
Chair of the WIAS Annual Conference	2018-2019
Chair of the WAPS council	2019-2020

Education and training total:	**49.6 ECTS**

[1] With the educational activities listed, the PhD candidate has complied with the educational requirements set by the graduate school Wageningen Institute of Animal Sciences (WIAS) of Wageningen University & Research, which comprises a minimum of 30 ECTS (European Credit Transfer and accumulation System). One ECTS equals a study load of 28 hours.

Colophon

The research described in this thesis was financed by DairyNL (ZuivelNL; organization of the Dutch dairy supply chain, The Hague, the Netherlands) and the Dutch Ministry of Agriculture, Nature and Food Quality (LNV, The Hague) as part of the research program One Health for Food (1H4F, The Hague).

Cover design by Fenna Schaap

Printed by ProefschriftMaken on 100% recycled paper